The 25th EURL-AR Proficiency Test Salmonella, Campylobacter and genotypic characterisation 2018

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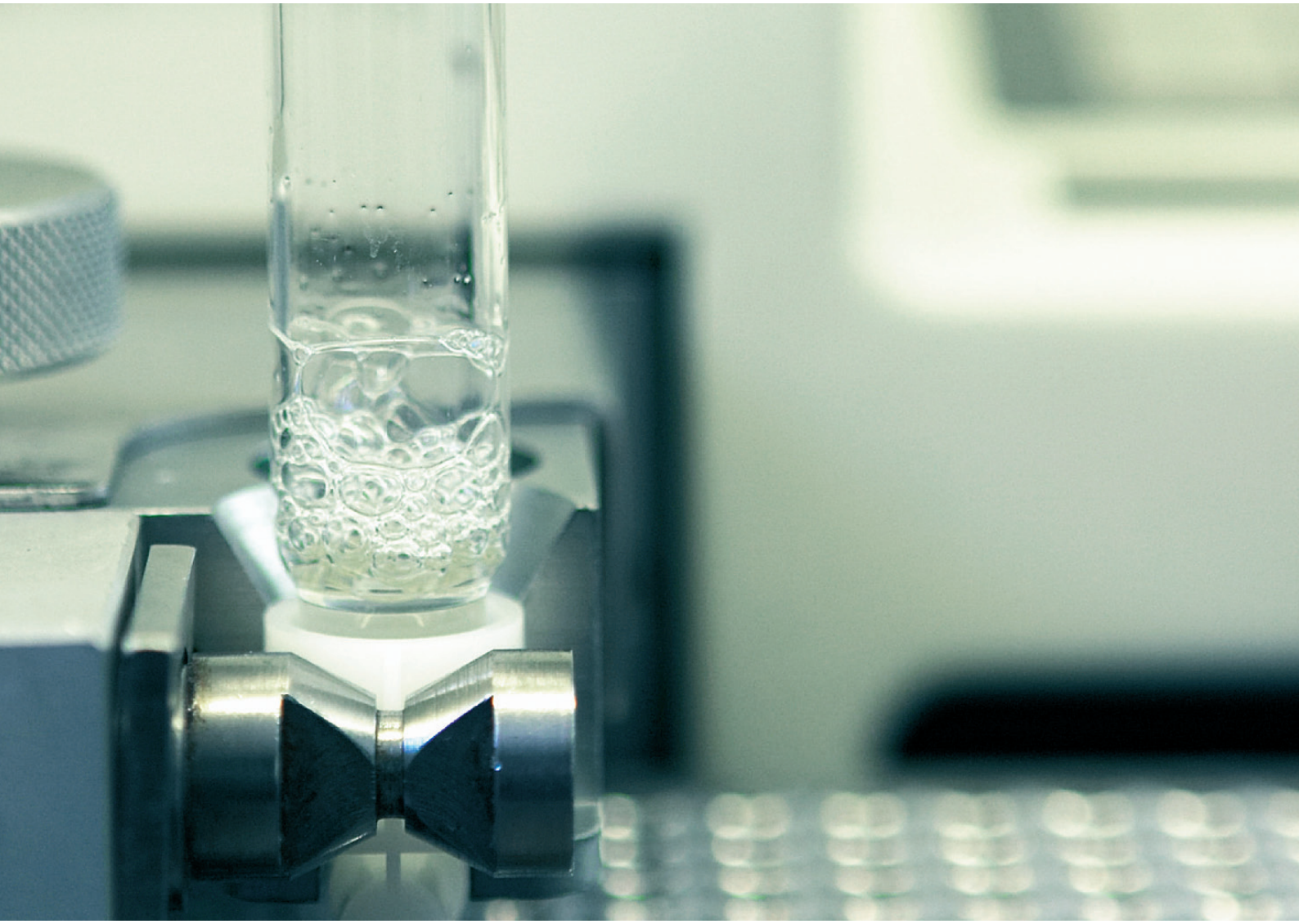

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THE 25th EURL-AR PROFICIENCY TEST
Salmonella, Campylobacter and genotypic characterisation - 2018

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1. Introduction

This report describes and summarises results of the twenty-fifth proficiency test trial conducted by the National Food Institute (DTU Food) as the EU Reference Laboratory for Antimicrobial Resistance (EURL-AR). This proficiency test focuses on antimicrobial susceptibility testing (AST) of *Salmonella* and *Campylobacter* and is the twelfth External Quality Assurance System (EQAS) conducted for these microorganisms (the first was EQAS 2006). In addition, the proficiency test includes categorisation of the relevant *Salmonella* strains as presumptive ESBL-, AmpC- and carbapenemase-producing organisms, and identification of the *Campylobacter* species as either *C. jejuni* or *C. coli*.

In addition, for the tenth time, an optional element was included, consisting of genotypic characterisation of antimicrobial resistance genes by PCR and/or sequencing. This optional component included characterisation of genes encoding ESBL-, AmpC- and carbapenemases in the *Salmonella* test strains.

This EQAS aims to: i) monitor the quality of AST results produced by National Reference Laboratories (NRL-AR), ii) identify laboratories which may need assistance to improve their performance in AST, and iii) determine possible topics for further research or collaboration.

In reading this report, the following important considerations should be taken into account:

1) Expected results were generated by performing Minimum Inhibitory Concentration (MIC) determinations for all test strains in two different occasions at the Technical University of Denmark, National Food Institute (DTU Food). These results were then verified by the Centers for Disease Control and Prevention, Georgia, US (*Salmonella*) and the United States Food and Drug Administration (FDA), Centre for Veterinary Medicine, Maryland, US (*Salmonella* and *Campylobacter*). Finally, a MIC determination was performed at DTU Food after preparation of the agar stab culture/charcoal swab for shipment to participants to confirm that the vials contained the correct strains with the expected MIC values.

2) Evaluation is based on interpretations of AST values determined by the participants. This is in agreement with the method used by Member States (MS) to report AST data to the European Food Safety Authority (EFSA), and complies with the main objective of this EQAS, i.e. to evaluate and improve the comparability of surveillance data on antimicrobial susceptibility of *Salmonella* and *Campylobacter* reported to EFSA by different laboratories, as stated in the protocol.

3) The EURL-AR network agreed on setting the acceptable deviation level for laboratory performance on AST to 5%. For the optional genotypic characterisation, no specific acceptance level has been set.

Evaluation of a result as “deviating from the expected interpretation” should be carefully analyzed in a self-evaluation procedure performed by the participant including also considerations related to any corrective actions introduced in the laboratory. Note that it is not considered a mistake to obtain a one-fold dilution difference in the MIC of a specific antimicrobial when testing the same strains since methods used for MIC determination have limitations. If, however, the expected MIC is close to the breakpoint value for categorising the strain as susceptible or resistant, a one-fold dilution difference - which is acceptable - may result in two different interpretations, i.e. the same strain can be categorised as susceptible or resistant. This result may be evaluated as correct based on the MIC-value produced but incorrect when the evaluation is based on the interpretation of the MIC value. This report is based on evaluation of AST interpretations,

therefore some participants may find their results classified as incorrect even though the actual MIC they reported is only a one-fold dilution away from the expected MIC. In these cases, the participants should be confident about the good quality of their performance of AST by MIC. In the organization of the EQAS, we try to avoid these situations by choosing test strains with MIC values distant from the cut offs for resistance, which is not always feasible for all strains and all antimicrobials. Therefore, the EURL-AR network unanimously established in 2008 that if there are less than 75% correct results for a specific strain/antimicrobial combination, the reasons for this situation must be further examined and, on selected occasions explained in details case by case, these results may subsequently be omitted from the evaluation report.

This report is approved in its final version by a technical advisory group composed by competent representatives from all NRL-ARs. This group meets annually at the EURL-AR workshop.

All conclusions presented in this report are publically available. Participating laboratories are identified by codes and each code is known only by the corresponding laboratory. The full list of laboratory codes is confidential and known only by relevant representatives of the EURL-AR and the EU Commission.

The EURL-AR is accredited by DANAK as provider of proficiency testing (accreditation no. 516); working with zoonotic pathogens and indicator organisms as bacterial isolates (identification, serotyping and antimicrobial susceptibility testing).

2. Materials and Methods

2.1 Participants in EQAS 2018

A pre-notification (Appendix 1) to announce the EURL-AR EQAS on AST of *Salmonella* and *Campylobacter* was distributed on the 28th August 2018 by e-mail to the 45 laboratories in the EURL-AR-network including all EU countries and Iceland, North Macedonia, Norway, Serbia, Switzerland and Turkey. All EU MS as well as Iceland, Norway, and Switzerland were represented as participants for both *Salmonella* and *Campylobacter.* FYROM was represented as a participant for *Salmonella*. In addition to the AST of *Salmonella* and *Campylobacter*, an optional genotypic characterisation by PCR/sequencing of antimicrobial resistance genes of the ESBL-, AmpC- and carbapenemase-producing *Salmonella* test strains was offered. Eleven laboratories participated in the optional genotypic characterisation of the ESBL-, AmpC- and carbapenemase-producing *Salmonella* test strains (see Appendix 2).

In addition to the MS NRLs, six additional laboratories were included as participants in the EQAS; one from each of the following countries: North Macedonia, Iceland, the Netherlands, Norway, Spain, and Switzerland. These were invited to take part in the EQAS 2018 on the basis of their participation in previous EQAS iterations and/or affiliation to the EU network. These laboratories were charged a fee for their participation in the EQAS, whereas the NRLs from EU Member States participated free of charge.

The results evaluated and presented in this report (N=32; Appendix 2) are from the NRLs designated by the MS (n=29) and NRLs in affiliated non-MS (n=4) (North Macedonia, Iceland, Norway, and Switzerland). Figure 1 illustrates the 32 participating countries.

In total, this report evaluates 32 sets of results from the *Salmonella* AST component, 31 sets of results from the *Campylobacter* AST component and eleven sets of results in relation to the optional genotypic characterisation.

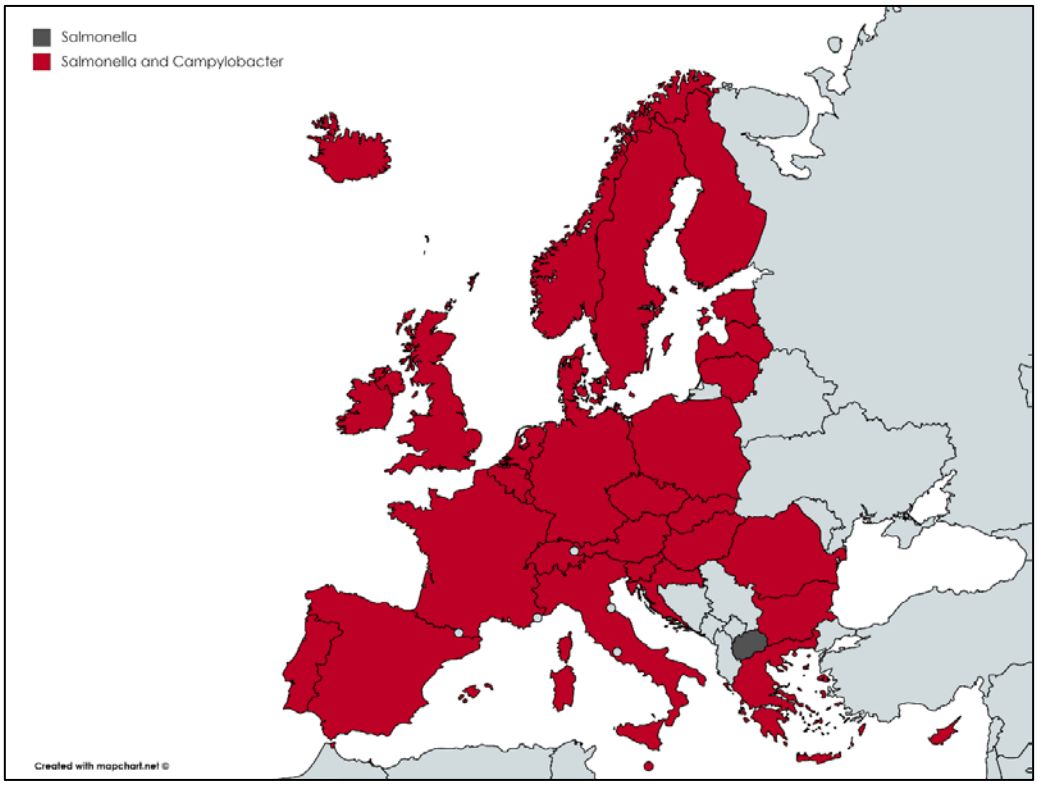

Figure 1: Participating countries that performed antimicrobial susceptibility testing of *Salmonella* and *Campylobacter* in 2018.

Results from the laboratories not designated by the MS but enrolled in the EQAS are not further presented or evaluated in this report.

2.2 Strains

Eight *Salmonella* strains and eight *Campylobacter* strains were selected for this trial among isolates from the strain collection at DTU Food on the basis of antimicrobial resistance profiles and MIC values. For quality assurance purposes, one strain per bacterial species has been included in all EQAS iterations performed to date, representing an internal control.

Prior to distribution of the strains, DTU Food performed AST on the *Salmonella* and *Campylobacter* strains and the AST profiles were verified by the Centers for Disease Control and Prevention (CDC), Georgia, US (*Salmonella*) and the United States Food and Drug Administration (FDA), Centre for Veterinary Medicine, Maryland, US (*Salmonella* and *Campylobacter*). When MIC-values were not in agreement but varied +/- one dilution-step, the value obtained by DTU Food was selected as the reference value. The obtained MIC values served as reference for the test strains (Appendix 3a and 3b). Results for the following antimicrobials were not verified by CDC or FDA for *Salmonella:* cefepime, cefotaxime, cefotaxime/clavulanic acid, ceftazidime, ceftazidime/clavulanic acid, colistin, ertapenem, imipenem, temocillin, tigecycline and trimethoprim, and results for the following antimicrobials were not verified by FDA for *Campylobacter*: streptomycin.

Reference strains *Escherichia coli* CCM 3954 (ATCC 25922) and *Campylobacter jejuni* CCM 6214 (ATCC 33560) had been forwarded to all participating laboratories when they were new participants with instructions to store and maintain them for quality assurance purposes

and future EQAS trials. Moreover, the EURL-AR has distributed *Campylobacter coli* (2012-70-443-2) and *Acinetobacter baumannii* (2012-70-100-69) for the purpose of performing internal control of the method.

2.3 Antimicrobials

The antimicrobials tested in this EQAS are listed in the protocol (Appendix 4b).

The antimicrobials tested correspond to the panel of antimicrobials listed in Decision 2013/652/EU.

The method applied for the AST was the ISO standard, ISO 20776-1 "Clinical laboratory testing and *in vitro* diagnostic test system – Susceptibility testing of infectious agents and evaluation of performance of antimicrobial susceptibility test devices", and, in addition, the following guidelines/standards from the Clinical and Laboratory Standards Institute (CLSI) were applied: Document M7-A11 (2018) "Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Eleventh Edition"; document M100, 28th ed. (2018) "Performance Standards for Antimicrobial Susceptibility Testing", document VET01 (2018) "Performance Standards for Antimicrobial Disk and Dilution Susceptibility Tests for Bacteria Isolated From Animals" – Fifth Edition; and document VET06 (2017) "Methods for Antimicrobial Susceptibility Testing of Infrequently Isolated or Fastidious Bacteria Isolated from Animals" – First Edition.

MIC results were interpreted by using the interpretative criteria listed in Decision 2013/652/EU. Where cut-off values were not available, the list of interpretative criteria was supplemented either with CLSI-interpretative criteria or with tentative values as described in the protocol (Appendix 4). No interpretative criteria were available for cefepime. Results for presumptive beta-lactam resistance mechanisms were interpreted according to the most recent EFSA recommendations also included as an appendix in the EQAS protocol (Appendix 4).

The selection of antimicrobials used in the trial for *Salmonella* were: ampicillin (AMP), azithromycin (AZI), cefepime (FEP), cefotaxime (FOT), cefotaxime/clavulanic acid (FOT/Cl), cefoxitin (FOX), ceftazidime (TAZ), ceftazidime/clavulanic acid (TAZ/Cl), chloramphenicol (CHL), ciprofloxacin (CIP), colistin (COL), ertapenem (ERT), gentamicin (GEN), imipenem (IMI), meropenem (MER), nalidixic acid (NAL), sulfonamides (sulfamethoxazole) (SMX), tetracycline (TET), tigecycline (TGC), temocillin (TRM) and trimethoprim (TMP).

Minimum Inhibitory Concentration (MIC) determination of the *Salmonella* test strains was performed using the Sensititre system (EUVSEC and EUVSEC2) from Trek Diagnostic Systems Ltd, UK.

For *Campylobacter* the following antimicrobials were included: ciprofloxacin (CIP), erythromycin (ERY), gentamicin (GEN), nalidixic acid (NAL), streptomycin (STR), and tetracycline (TET).

MIC determination for the *Campylobacter* testing was performed using the Sensititre systems (EUCAMP2) from Trek Diagnostic Systems Ltd, UK. Participants of the *Campylobacter* EQAS were additionally requested to identify the species of the *Campylobacter* spp. as either *C. jejuni* or *C. coli*.

2.4 Distribution

On 16 October 2018, bacterial strains in agar stab cultures (*Salmonella* spp.) or charcoal swabs in transport media (Stuarts) (*Campylobacter* spp.) together with a welcome letter (Appendix 4a) were dispatched in double pack containers (class UN 6.2) to the participating laboratories. The shipment (UN3373, biological substances category B) was sent according to International Air Transport Association (IATA) regulations.

2.5 Procedure

Protocols and all relevant information were uploaded on the EURL-AR website (http://www.eurl-ar.eu). thereby EQAS participants could access necessary information at any time.

Participants were instructed to subculture charcoal swabs immediately and store the agar stabs at 4ºC (dark) until performance of AST. Information related to the handling of the test strains and reference strains (Appendix 4b, c, d, e) was made available.

The participants were instructed to apply the interpretative criteria listed in the protocol (Appendix 4). Instructions for interpretation of AST results allowed for categorisation of strains as resistant or susceptible. Categorisation as 'intermediate' was not accepted.

The EURL-AR is aware that there are two different types of interpretative criteria of results, i.e clinical breakpoints and epidemiological cut-off values. The terms 'susceptible', 'intermediate' and 'resistant' should be reserved for classifications made in relation to the therapeutic application of antimicrobial agents. When reporting data using epidemiological cut-off values, bacteria should be reported as 'wild-type' or 'non-wild-type' (Schwarz *et al*., 2010). To simplify the interpretation of results, throughout this report, we will maintain the terms susceptible and resistant, even if referring to wild-type and non-wild-type strains, respectively.

As regards the method for performing the antimicrobial susceptibility testing, the protocol referred to Decision 2013/652/EU and instructed participants to perform the international reference method for antimicrobial susceptibility testing. I.e. dilution methods performed according to the methods described by the European Committee on Antimicrobial Susceptibility Testing (EUCAST) and the Clinical and Laboratory Standards Institute (CLSI), accepted as the international reference method (ISO standard 20776-1:2006).

A mandatory part of the proficiency test was to detect ESBL-, AmpC- and carbapenemase-producing strains and interpret results according to the most recent EFSA recommendations as described in the protocol.

Results for QC reference strains were MIC values for the reference strains *E. coli* (ATCC 25922) and *C. jejuni* (ATCC 33560). The results were evaluated towards the quality control ranges according to the relevant guidelines; i.e. the CLSI documents VET06 (2017) or M100S, 28th ed. (2018) (Appendix 5).

For the optional genotypic characterisation of the genetic basis for resistance to extended-spectrum cephalosporins and carbapenems in the *Salmonella* test strains, participating laboratories were invited to report the genes. For the test strains included in this component of the EQAS, the organizers, decided to include $bla_{\text{TEM-1}}$ (encoding TEM-beta-lactamases) as an expected gene. The genes listed in the table in the protocol (Appendix 4b) were included in the test. Identification of additional genes not listed in the protocol was not evaluated by the database. The results were evaluated based on the identified genes and variants.

The participating laboratories were encouraged to use their own laboratory's method(s) for the genotypic characterisation. The expected results for this component of the EQAS were obtained by whole-genome-sequencing and subsequent analysis using the ResFinder 3.0 platform available at http://cge.cbs.dtu.dk/services/ResFinder/. The positive identification of genes was not verified elsewhere.

All participating laboratories were invited to enter the obtained results into an electronic record sheet at the EURL-AR web-based database through a secured individual login and password.

In addition, participants were encouraged to complete an evaluation form available at the EURL-AR database with the aim to improve future EQAS trials.

The database was finally closed and evaluations were made available to participants on February 7, 2019. After this date, the participants were invited to login to retrieve an individual, database-generated report which contained an evaluation of the submitted results including possible deviations from the expected interpretations. Deviations in interpretation (resistant or susceptible) were categorised as 'incorrect', as were also deviations concerning confirmation of an isolate as extended spectrum beta-lactamase- (ESBL-), AmpC- or carbapenemase-producer.

3. Results

The participants were asked to report results, i.e. MIC values and the categorisation as resistant or susceptible. Only the categorisation was evaluated, whereas the MIC values were used as supplementary information.

3.1 Data omitted from the report

As mentioned in the introduction, the EURL-AR network established that data should be examined and possibly omitted from the general analysis if there are less than 75% correct results based on strain/antimicrobial combination (see Appendix 7a and 7b for an overview of correct/incorrect results). In the present EQAS this occurred in two cases which have been examined and consequently omitted from the analysis; **1)** S-13.1/ceftazidime (expected interpretation was 'resistant', however, 37% of the submitted results (from 15 laboratories) found the strain susceptible in either panel 1 or panel 2. All but two of the deviating interpretations were based on MIC values one step from the expected; **2)** S-13.7/cefoxitin (expected interpretation was 'susceptible'), however, 35% found the strain resistant to cefoxitin. All of the deviating interpretations were based on MIC values within one step from the expected.

For *Campylobacter*, no problems specific for a strain/antimicrobial-combination were identified.

3.2 Methods

Results obtained by broth microdilution were accepted and evaluated. For both the *Salmonella* and the *Campylobacter* trial, all 32 and 31 laboratories, respectively, reported results obtained by broth microdilution.

With the aim to conclude on the strains' presumptive ESBL-, AmpC- and carbapenemase phenotype, two panels of antimicrobials were included in the testing of the *Salmonella* strains as also specified in the EU regulation 2013/652/EU. The test strains found resistant to cefotaxime, ceftazidime or meropenem on the first panel (see 2013/652/EU, Table 1) were additionally tested on the second panel (see 2013/652/EU, Table 4) according to the protocol indications.

3.3 Deviations, overall

The list of deviations is presented in Appendix 8a and 8b. Figure 2 shows the total percentage of deviations from the expected results of AST performed by participating laboratories. Overall, the deviation levels in 2018 are acceptable for both the *Salmonella* and the *Campylobacter* trials.

The internal control strains mainly followed the trend in deviation level of the different EQAS trials (Figure 2).

3.3.1 *Salmonella* trial

For the *Salmonella* strains, 99.0% of the AST results were correct. The number of AST's performed and the percentage of correct results for the individual strains in the EQAS, are listed

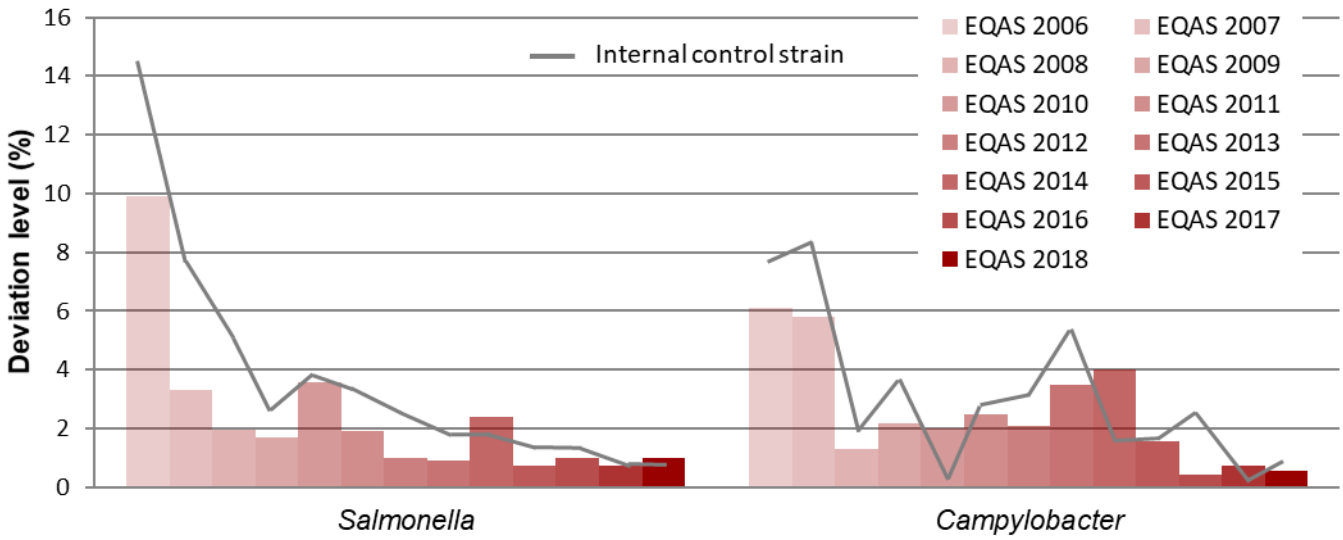

Figure 2: A comparison between the EURL-AR EQAS's since 2006, showing the total percentage of deviations for antimicrobial susceptibility testing performed by participating laboratories.

in Table 1. Variations of obtained correct results ranged from 96.3 to 99.8%. Table 2 illustrates the percentage of correct AST per antimicrobial by bacterial species. The level of correct AST was at 94.5% (cefoxitin) or above, for all the *Salmonella* test strains. For cefoxitin, four of the seven deviations could be attributed to one laboratory (#19) for which all incorrect interpretations were based on an obtained MIC value that was equal to the expected MIC value which was incorrectly interpreted as resistant.

ESBL/AmpC/carbapenemase-producing *Salmonella* test strains

Confirmation of beta-lactamase production is a mandatory component of this EQAS.

According to the protocol, which was based on the EFSA recommendations, the confirmatory test for ESBL-, AmpC-, and carbapenemase-producing isolates requires use of both cefotaxime (FOT) and ceftazidime (TAZ) alone and in combination with a β-lactamase inhibitor. The MIC value for either antimicrobial agent (FOT or TAZ) tested in combination with clavulanic acid should be compared to the corresponding MIC when tested alone. Synergy is defined for one or both cephalosporins if a ≥ 3-dilution-step difference is observed between the two MIC values (i.e. if the FOT:CTX/Cl or TAZ:TAZ/Cl ratio ≥8) (CLSI M100S Table 2A; Enterobacteriaceae). Participants were instructed to test strains to use the second panel of antimicrobials to test strains presenting resistance to cefotaxime (FOT), ceftazidime (TAZ or meropenem (MERO) on panel 1.

The classification of the phenotypic results was based on the most recent EFSA recommendations as indicated in the protocol (Appendix 4).

In this EQAS, all laboratories uploaded results for the strains to be tested on panel 2.

Table 3 presents that the strains S-13.1, S-13.4, S-13.7 and S-13.8 were ESBL producers, and strain S-13.6 was a carbapenemase producer. For strain S-13.7, some laboratories (34%) obtained an MIC value for cefoxitin at 16 and consequently reported a categorisation as 'resistant'. As mentioned above in paragraph

Table 1. The number of AST performed and the percentage of correct results for each strain of *Salmonella* (panel 1 and panel 2) and *Campylobacter*.

EQAS 2018 – *Salmonella*			EQAS 2018 – *Campylobacter*		
Test strain	AST in total	% correct	Test strain	AST in total	% correct
S-13.1	590	99.8	C-13.1 (*C. jejuni*)	186	98.9
S-13.2	445	99.8	C-13.2 (*C. coli*)	186	99.5
S-13.3	448	99.1	C-13.3 (*C. coli*)	180	98.9
S-13.4	655	99.2	C-13.4 (*C. jejuni*)	186	100.0
S-13.5	447	98.9	C-13.5 (*C. jejuni*)	186	100.0
S-13.6	650	96.3	C-13.6 (*C. jejuni*)	184	98.4
S-13.7	623	99.7	C-13.7 (*C. coli*)	186	100.0
S-13.8	655	99.5	C-13.8 (*C. jejuni*)	186	100.0

Table 2: Percentage of correct antimicrobial susceptibility tests per antimicrobial by microorganism.

Antimicrobial	*Salmonella*	*Campylobacter*
Ampicillin	99.6	-
Azithromycin	99.5	-
Cefotaxime	98.3	-
Cefoxitin	94.5	-
Ceftazidime	99.4	-
Chloramphenicol	99.6	-
Ciprofloxacin	98.8	99.6
Colistin	98.8	-
Ertapenem	99.4	-
Erythromycin	-	99.2
Gentamicin	100.0	100.0
Imipenem	96.9	-
Meropenem	98.8	-
Nalidixic acid	100.0	99.2
Streptomycin	-	100.0
Sulphonamides	97.6	-
Temocillin	99.2	-
Tetracycline	100.0	98.8
Tigecycline	100.0	-
Trimethoprim	99.2	-

3.1, the results related to the strain/antimicrobial combination were disregarded, consequently also the results in relation to categorisation as ESBL-phenotype or AmpC/ESBL phenotype were disregarded.

In total, the categorisation as ESBL-, AmpC- or carbapenemase-producer for the eight strains was correct in 218 out of 224 reported results. Of the results that were considered incorrect (N=6), three could be attributed to laboratory #64. The MIC results obtained by laboratory #64 for S-13.4 and S-13.5, however, do not indicate any reason for the incorrect ESBL-, AmpC- or carba-categorisation. The laboratory reported that the task was overlooked when the EQAS was performed, and consequently the results were submitted in spite of missing background information.

3.3.2 *Campylobacter* trial

For the *Campylobacter* strains, 99.5% of AST results were correct. Table 1 presents that the variation between the strains in the obtained correct results ranged from 98.9 to 100% and Table 2 illustrates that the percentage of correct AST per antimicrobial were all above 98.8%.

The participants were requested to identify the *Campylobacter* species as *C. jejuni* or *C. coli*. All 31 laboratories delivered in total 248 results of which all were in accordance with the expected.

3.4 Deviations by laboratory

Figure 3 and 4 illustrate the percentage of deviations for each participating laboratory. The laboratories are ranked according to their performance determined by the percentage of deviating results in the antimicrobial susceptibility tests.

Table 3: Overview of ESBL-, AmpC- and carbapenemase-producing *Salmonella* test strains and proportion of laboratories that obtained the expected result; number and percentages of laboratories which correctly detected and confirmed the ESBL-, AmpC- and carbapenemase-producing *Salmonella* strains.
Fields shaded in grey with numbers in *italics* indicate an unexpected result.
Grey text indicates results that is disregarded for further analysis.

Strain code		S -13.1	S-13.4	S-13.6	S-13.7*	S-13.8
ESBL/AmpC/carbapenemase-encoding genes harboured in the test strain		***bla*$_{CTX-M-8}$**	***bla*$_{CTX-M-9}$ *bla*$_{TEM-1B}$**	***bla*$_{OXA-48}$**	*bla*$_{SHV-12}$ *bla*$_{CTX-M-15}$	***bla*$_{CTX-M-3}$ *bla*$_{TEM-1B}$**
ESBL-, AmpC- and carbapenemase-producing strain – expected results		ESBL	ESBL	carbapenemase	ESBL*	ESBL
Obtained results	Confirmed ESBL-producer	32/32 (100%)	31/32 (97%)	-	23/32 (72%)	32/32 (100%)
	Confirmed ESBL + AmpC-producer	-	-	-	*9/32 (28%)*	-
	Confirmed AmpC-producer	-	-	-	-	-
	Confirmed carbapenemase-producer	-	-	29/32 (91%)	-	-
	Confirmed other phenotype	-	-	*1/32 (3%)*	-	-
	Not ESBL-, AmpC- or carbapenemase-producing	-	*1/32 (3%)*	*2/32 (6%)*	-	-

* Results from strain S-13.7 were disregarded for further analysis in this report due to the fact that for a number of laboratories (n=9) the cefoxitin MIC value indicated an ESBL + AmpC producer even if the MIC-value was within the method acceptance limit (± one one-fold dilution step).

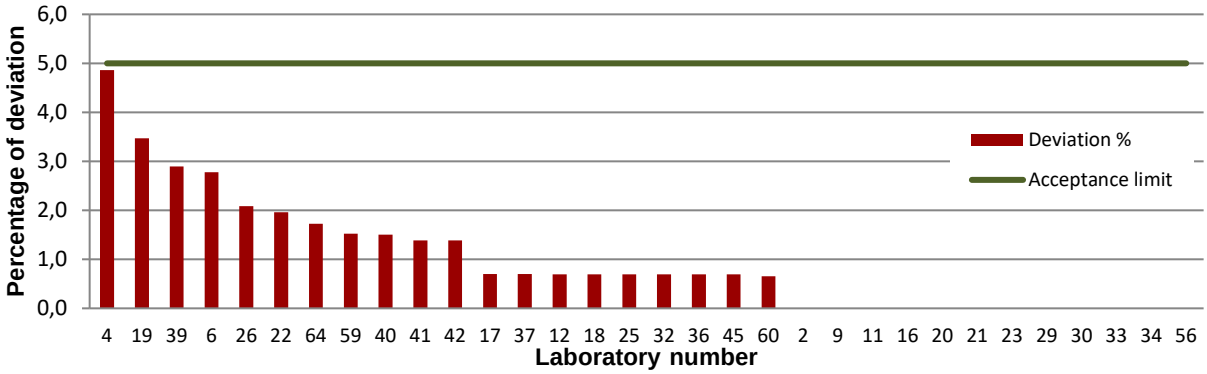

Figure 3: Individual participants' deviations in percent of their total number of *Salmonella* AST's.

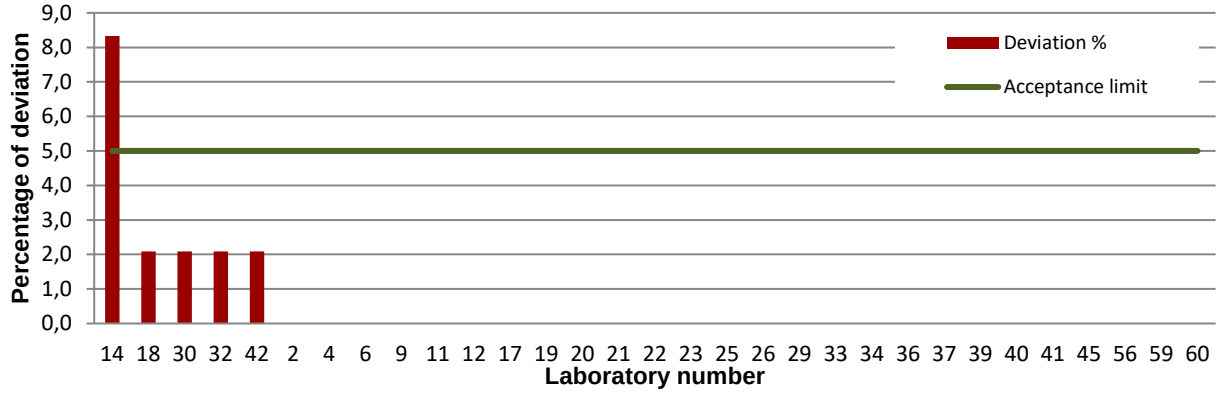

Figure 4: Individual participants' deviations in percent of their total number of *Campylobacter* AST's.

Table 4 Obtained values for AST of *E. coli* ATCC 25922. AMP; ampicillin, FEP; cefepime FOT; cefotaxime, FOX; cefoxitin, TAZ; ceftazidime, CHL; chloramphenicol, CIP; ciprofloxacin, COL; colistin, ERT: ertapenem, GEN; gentamicin, IMI; imipenem, MER; meropenem, NAL; nalidixic acid, SMX; sulphonamides, TET; tetracycline, TGC; tigecycline, TMP; trimethoprim.

MIC determination *E. coli* ATCC 25922			
Antimicrobial	Proportion outside QC range	Obtained values in MIC steps (min/max)	
		Below lower QC limit	Above upper QC limit
Panel 1, AMP	0/32 (0%)	-	-
Panel 1, FOT	1/31 (3%)	-	1 step
Panel 1, TAZ	0/32 (0%)	-	-
Panel 1, CHL	0/32 (0%)	-	-
Panel 1, CIP	0/32 (0%)	-	-
Panel 1, COL	0/32 (0%)	-	-
Panel 1, GEN	1/32 (3%)	2 steps	-
Panel 1, MER	0/32 (0%)	-	-
Panel 1, NAL	0/32 (0%)	-	-
Panel 1, SMX	0/31 (0%)	-	-
Panel 1, TET	0/32 (0%)	-	-
Panel 1, TGC	1/32 (3%)	-	1 step
Panel 1, TMP	2/32 (6%)	1 step	-
Panel 2, FEP	0/28 (0%)	-	-
Panel 2, FOT	0/28 (0%)	-	-
Panel 2, FOX	0/29 (0%)	-	-
Panel 2, TAZ	0/29 (0%)	-	-
Panel 2, ERT	0/29 (0%)	-	-
Panel 2, IMI	0/29 (0%)	-	-
Panel 2, MER	0/29 (0%)	-	-

Table 5 Obtained values for AST of *C. jejuni* ATCC 33560. CIP; ciprofloxacin, ERY; erythromycin, GEN; gentamicin, NAL; nalidixic acid, TET; tetracycline.

MIC determination *C. jejuni* ATCC 33560			
Antimicrobial	Proportion outside QC range	Obtained values in MIC steps (min/max)	
		Below lower QC limit	Above upper QC limit
CIP	0/30 (0%)	-	-
ERY	0/29 (0%)	-	-
GEN	3/30 (10%)	1 step	-
NAL	0/30 (0%)	-	-
TET	1/30 (3%)	-	1 step

3.4.1 *Salmonella* trial

All 32 participating laboratories obtained a result within the acceptance limit (< 5% deviations) for the *Salmonella* strains. The maximum percentage of deviations was at 4.9%, presenting acceptable result across the EURL-AR network.

3.4.2 *Campylobacter* trial

In the Campylobacter trial, most laboratories performed very well. Applying the 5% acceptance threshold, 30 of 31 participating laboratories performed acceptably, with 26 laboratories having no deviations (Figure 4).

One laboratory presented a deviation level at 8.3%, i.e. above the 5% acceptance level (#14).

3.5 Deviations by reference strains

In the following section, deviations are defined as results of antimicrobial susceptibility tests on the reference strain that are outside the quality control (QC) acceptance intervals (Appendix 5).

Obtained values from the participants' testing of the QC strains are listed in Appendix 6a and 6b, and in Tables 4 and 5. For the *Salmonella* and *Campylobacter* trial, 32 and 28 laboratories, respectively, uploaded data from testing of the relevant QC strain.

Appendix 6a presents the results for the reference strain *E. coli* ATCC 25922. Four laboratories produced in total five values outside the acceptable range. Table 4 illustrates the obtained results which are fully presented in Appendix 6a.

Table 5 presents the proportion of the laboratories submitting AST-results for the *C. jejuni* reference strain ATCC 33560 with results below or above the acceptable range. Four deviations were observed from four different laboratories.

3.6 Genotypic characterisation

For the optional genotypic characterisation of the ESBL-, AmpC-, and carbapenemase-

producing *Salmonella* test strains, 11 laboratories participated. In Appendix 9, information is presented on detected genes, primers used, and references for the method used. Five laboratories reported results based on whole genome sequencing of the ESBL-, AmpC- and carbapenemase-producing *Salmonella* whereas five laboratories indicated the use of various types of PCR to identify the relevant genes. The remaining laboratory applied both WGS and PCR for the detection of the ESBL-, AmpC- and carbapenemase-encoding genes. Table 6 indicates the obtained results, both on family and variant level. Moreover, Figure 5 indicates that two discordant results related to one gene and one variant were submitted by two different laboratories. These were related to the *bla*$_{DHA}$ gene and a variant of the *bla*$_{CTX-M}$ gene.

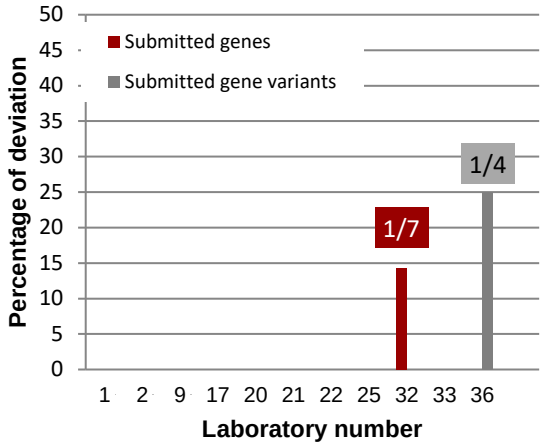

Figure 5: Deviations as a percentage of the total number of results (i.e. detected genes) reported by each participant.

Table 6: Results from the participation of eleven laboratories in the optional genotypic characterisation component of the EQAS.

Strain code	Expected gene	Proportion of correct results (family level)	Proportion of correct results (variant level)	Unexpected genes/variants identified
S-13.1	*bla*$_{CTX-M-8}$	10/10 (100%)	8/8 (100%)	*bla*$_{DHA}$ (n=1)
S-13.4	*bla*$_{CTX-M-9}$	11/11 (100%)	8/8 (100%)	
	bla$_{TEM-1/1B}$	9/9 (100%)	7/7 (100%)	
S-13.6	*bla*$_{OXA-48}$	11/11 (100%)	10/10 (100%)	
S-13.7	*bla*$_{SHV-12}$	11/11 (100%)	8/8 (100%)	
	bla$_{CTX-M-15}$	11/11 (100%)	9/9 (100%)	
S-13.8	*bla*$_{CTX-M-3}$	11/11 (100%)	7/8 (88%)	*bla*$_{CTX-M-15}$ (n=1)
	bla$_{TEM-1/1B}$	9/9 (100%)	8/8 (100%)	

4. Discussion

The number of participating laboratories was at comparable levels in 2018 as in 2017; for the *Salmonella* EQAS, 31 and 32 participated in 2017 and 2018, respectively, and for *Campylobacter* EQAS, 30 and 31 participated in 2017 and 2018. This allows for a fair comparison between the two EQAS periods for both organisms.

As also specified in the EU regulation 2013/652/EU, all participants in the present EQAS performed AST by broth microdilution.

This 2018 proficiency test was the fifth possibility of testing *Salmonella* and *Campylobacter* strains with the panels designed to follow the requirements of Decision 2013/652/EU.

4.1 *Salmonella* trial

Overall, the percentage of correct antimicrobial susceptibility test results of *Salmonella* was

99.0%. All (n=32) participants obtained satisfactory results according to the level of acceptance (<5% deviation) (Figure 3). When comparing between the antimicrobials, the testing of cefoxitin appeared to cause most problems (94.5% correct results).

As indicated in Figure 2, the overall quality of the results in the 2018-EQAS would appear to be at the same high level as in 2017, also, when comparing results obtained from testing the internal control strain a steady and very good quality of *Salmonella* AST results was observed.

Based on these results, follow-up has not been necessary and none of the laboratories was defined as outlier.

For the *E. coli* reference strain, the obtained results were in general in agreement with the CLSI recommendations.

Follow up on previous EQAS results was not relevant as no laboratories had deviation levels for the AST results above the acceptance limit in EQAS 2017.

ESBL/AmpC/carbapenemase-producing *Salmonella* test strains

The phenotypic detection of ESBL-, AmpC- and carbapenemase-producing microorganisms remains to be important and is a mandatory part of this EQAS.

Of the five *Salmonella* test strains relevant for this component of the EQAS (S-13.1, S-13.4, S-13.6, S-13.7, and S-13.8), one was a carbapenemase producer (S-13.6) and four were ESBL-producers. One of the strains (S-13.7) was problematic due to an MIC for cefoxitin at the breakpoint causing the strain to be categorised as an AmpC+ESBL-producer by 9 of 32 laboratories whereas the expected category was 'ESBL-producer'. Therefore, this strain was therefore disregarded for the evaluation.

The testing and interpretation of results for the ESBL- and carbapenemase-producing strains appeared not to cause difficulties. One laboratory (#64), however, obtained three of the total number of six deviations though these incorrect categorisations could not be explained by the submitted MIC results. Follow-up has been done directly with the laboratory to identify what might be done to improve the interpretation performance.

Even if no acceptance limit has been defined for this component of the EQAS, the overall result appears satisfactory.

4.2 *Campylobacter* trial

For the *Campylobacter* component of this year's EQAS, 31 laboratories submitted results leading to an overall percentage of correct AST results at 99.5%. The performance varied from no deviations to up to 8.3% deviations, with 30 laboratories performing satisfactorily according to the established acceptance range.

It appears that the level of deviations for the overall AST results is similar to the EQAS 2017, also in relation to the results obtained from testing the internal control strain (Figure 2).

One laboratory (#14) obtained deviation levels above 5%. For this laboratory, the four obtained deviations that caused the 8.3% deviation level were related to two strains (C-13.1 and C-13.6) and to the testing of nalidixic acid and tetracycline and indicate that a switch of strains may have happened.

All participating laboratories except one (#29) uploaded data for tests performed on the *C. jejuni* reference strain and the proportion of results within the acceptable range was 97.3%.

All four values outside the QC intervals were one step below or above the QC-limits. It is suggested that these values are monitored over time to ensure that the tests render a reliable result for the particular antimicrobial.

In 2017, one laboratory (#40) obtained AST results outside the acceptance limit (at 10.4%).

The EURL-AR followed-up directly with this laboratory and conveyed suggestions for improvements for performing AST's. This year, laboratory #40 obtained no deviations when performing *Campylobacter* AST.

4.3 Genotypic characterisation

One of the unexpected results, *bla*$_{DHA}$ reported by laboratory #32, was reported due to a transcription error (an error in the drop-down tab of EQAS data entry).

Genotypic characterisation of microorganisms has been applied in a number of laboratories and work is ongoing to improve laboratory detection of antimicrobial resistance genes, including identification of ESBL-, AmpC-, and carbapenemase-producing *Enterobacteriaceae*.

The optional genotypic characterisation is a supplementary component of this EQAS and should therefore be seen as an important possibility for the NRL-AR's to introduce or improve these methods in the laboratory. This year, eleven laboratories participated in the optional EQAS component and even though no acceptance limit has been defined, the 98.7% correct results (N=150) appears to be a very satisfactory result.

5. Conclusions

The goal of the EURL-AR EQAS is to have all participating NRLs performing antimicrobial susceptibility testing of *Salmonella* and *Campylobacter* with a deviation level below 5%. This year, this goal was reached for both *Salmonella* and *Campylobacter*.

Compared to the EQAS 2017, the performance of the NRL's in 2018 appears to be at the same high level for *Salmonella* AST's (99.0% in 2018 and 99.3% in 2017) as well as for *Campylobacter* (99.5% in 2018 and 99.3% in 2017) (Figure 2).

The test covering the identification of the phenotype of *Salmonella* test strains producing beta-lactamases of the ESBL-, AmpC, and carbapenemase type rendered six deviations (97.3% correct categorisations). This is a priority area within the EURL-AR activities, and the focus on identifying ESBL-, AmpC-, and carbapenemase-producing organisms is encouraged.

Eleven NRLs participated in the EQAS component consisting of genotypic testing of ESBL-, AmpC- and carbapenemase-producing *Enterobacteriaceae* presenting satisfactory results.

Finally, the EURL-AR is open to suggestions to improve future EQAS trials and invites the entire network to contribute with ideas for training courses and specific focus areas to expand the network's knowledge in antimicrobial resistance.

6. References

European Commission, 2013/652/EU: Commission Implementing Decision of 12 November 2013 on the monitoring and reporting of antimicrobial resistance in zoonotic and commensal bacteria.

Schwarz S, Silley P, Simjee S, Woodford N, van DE, Johnson AP & Gaastra W. (2010) Editorial: assessing the antimicrobial susceptibility of bacteria obtained from animals. J Antimicrob Chemother 65: 601-604

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G00-06-001/23.06.2017

EQAS 2018 FOR *SALMONELLA, CAMPYLOBACTER* AND OPTIONAL GENOTYPIC CHARACTERISATION

The EURL-AR announces the launch of another EQAS, thus providing the opportunity for proficiency testing which is considered an essential tool for the generation of reliable laboratory results of consistently good quality.

This EQAS consists of antimicrobial susceptibility testing of eight *Salmonella* isolates and eight *Campylobacter* isolates. Additionally, quality control (QC) strains *E. coli* ATCC 25922 (CCM 3954) and *C. jejuni* ATCC 33560 (CCM 6214) will be distributed to new participants.

It is the recipients' responsibility to comply with national legislation, rules and regulation regarding the correct use and handling of the provided strains and to possess the proper equipment and protocols to handle these strains.

This EQAS is specifically for NRL's on antimicrobial resistance (NRL-AR). Laboratories designated to be NRL-AR do not need to sign up to participate but are automatically regarded as participants. You may contact the EQAS-Coordinator if you wish to inform of changes in relation to your level of participation in compared to previous years. The EURL-AR will be able to cover the expenses for one parcel, only, per EU Member State. Therefore, countries with more than one laboratory registered on the EURL-AR contact-list will be contacted directly to confirm which laboratory will be included for participation free of charge.

The invitation to participate in the proficiency test is extended to additional participants besides official NRLs and to participants from laboratories which are involved in the network but are not designated NRLs (cost for participation will be 100 EUR).

TO AVOID DELAY IN SHIPPING THE ISOLATES TO YOUR LABORATORY

The content of the parcel is "UN3373, Biological Substance Category B": Eight *Salmonella* strains, eight *Campylobacter* and for new participants also the QC strains mentioned above. Please provide the EQAS coordinator with documents or other information that can simplify customs procedures (e.g. specific text that should be written on the proforma invoice). To avoid delays, we kindly ask you to send this information already at this stage.

TIMELINE FOR RESULTS TO BE RETURNED TO THE NATIONAL FOOD INSTITUTE

Shipment of isolates and protocol: The isolates will be shipped in October 2018. The protocol for this proficiency test will be available for download from the website (www.eurl-ar.eu).

Submission of results: Results must be submitted to the National Food Institute **no later than December 7th 2018** via the password-protected website.
Upon reaching the deadline, each participating laboratory is kindly asked to enter the password-protected website once again to download an automatically generated evaluation report.

EQAS report: A report summarising and comparing results from all participants will be issued. In the report, laboratories will be presented coded, which ensures full anonymity. The EURL-AR and the EU Commission, only, will have access to un-coded results. The report will be publicly available.

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Next EQAS: The next EURL-AR EQAS that we will have is on antimicrobial susceptibility testing of *E. coli*, staphylococci and enterococci which will be carried out in June 2019.

Please contact me if you have comments or questions regarding the EQAS.

Sincerely,

Susanne Karlsmose Pedersen
EURL-AR EQAS-Coordinator

Participant list

Salmonella	*Campylobacter*	Genotypic characterisation	Institute	Country
X	X	X	Austrian Agency for Health and Food Safety	Austria
X	X	-	Sciensano	Belgium
X	X	-	National Diagnostic and Research Veterinary Institute	Bulgaria
X	X	-	Croatian Veterinary Institut	Croatia
X	X	-	Veterinary Services	Cyprus
X	X	X	State Veterinary Institute Praha	Czech Republic
X*	X*	X	DTU National Food Institute	Denmark
X	X	-	Danish Veterinary and Food Administration, DVFA	Denmark
X	X	-	Estonian Veterinary and Food Laboratory	Estonia
X	X	-	Finnish Food Authority	Finland
X	-	-	Agence nationale de sécurité sanitaire ANSES - Fougères LERMVD	France
-	X	-	Agence nationale de sécurité sanitaire ANSES - Ploufragan - LERAP	France
X	X	X	Federal Institute for Risk Assessment	Germany
X	X	-	Veterinary Laboratory of Chalkis	Greece
X	X	-	Central Agricultural Office Veterinary Diagnostic Directorate	Hungary
X	X	-	University of Iceland	Iceland
X	X	X	Central Veterinary Research Laboratory	Ireland
X	X	X	Istituto Zooprofilattico Sperimentale delle Regioni Lazio e Toscana	Italy
X	X	X	Institute of Food Safety, Animal Health and Enviroment "BIOR"	Latvia
X	X	-	National Food and Veterinary Risk Assessment Institute	Lithuania
X	X	-	Laboratoire National de Santé	Luxembourg
X	X	-	Public Health Laboratory	Malta
X	X	X	Central Veterinary Institute of Wageningen UR	Netherlands
X*	X*	-	Food and Consumer Product Safety Authority (VWA)	Netherlands
X	X	X	Veterinærinstituttet	Norway
X	X	-	National Veterinary Research Institute	Poland
X	X	-	Laboratorio National de Investigacáo Veterinaria	Portugal
X	X	-	Institute for Hygiene and Veterinary Public Health	Romania
X*	X*	X*	Institute for Diagnosis and Animal Health	Romania
X	X	-	State Veterinary and Food Institute (SVFI)	Slovakia
X	X	-	National Veterinary Institute	Slovenia
X	X	X	Laboratorio Central de Sanidad, Animal de Algete	Spain
X*	-	-	Centro Nacional de Alimentación (AECOSAN)	Spain
X*	X*	-	VISAVET Health Surveillance Center, Complutense University	Spain
X	X	X	National Veterinary Institute, SVA	Sweden
X	X	-	Vetsuisse Faculty Bern, Institute of Veterinary Bacteriology	Switzerland
X	-	-	Faculty of Veterinary Medicine - Skopje	The former Yugoslav Republic of Macedonia
X*	X*	-	Agri-Food and Biosciences Institute (AFBI)	United Kingdom
X*	-	-	Public Health England	United Kingdom
X	X	-	Animal Plant Health Agency	United Kingdom

Designated NRL-AR by the compentent authority of the member state
Non-NRL-AR enrolled by the EURL-AR
Not a Member State of the EU
* Submitted results were not included in the current report (allows for one dataset per country, only)

Reference values (MIC-value and interpretation) - *Salmonella*

	Ampicillin AMP		Azithromycin AZI		Cefepime FEP		Cefotaxime FOT		Cefotaxime/clav F/C	F:F/C ratio	Cefoxitin FOX		Ceftazidime TAZ		Ceftazidime/clav T/C	T:T/C ratio	Chloramphenicol CHL		Ciprofloxacin CIP		Colistin COL		Ertapenem	
EURL 2018 S-13.1	>64	RESIST	8	SUSC	32	NA	64	RESIST	0.12	>=8	8	SUSC	4	RESIST	0.5	>=8	<=8	SUSC	0.03	SUSC	<=1	SUSC	<=0.015	SUSC
EURL 2018 S-13.2	>64	RESIST	>64	RESIST			<=0.25	SUSC					<=0.5	SUSC			<=8	SUSC	<=0.015	SUSC	<=1	SUSC		
EURL 2018 S-13.3	2	SUSC	4	SUSC			<=0.25	SUSC					<=0.5	SUSC			<=8	SUSC	<=0.015	SUSC	<=1	SUSC		
EURL 2018 S-13.4	>64	RESIST	8	SUSC	2	NA	16	RESIST	0.12	>=8	4	SUSC	1	SUSC	0.25	<8	<=8	SUSC	0.5	RESIST	<=1	SUSC	<=0.015	SUSC
EURL 2018 S-13.5	<=1	SUSC	4	SUSC			<=0.25	SUSC					<=0.5	SUSC			<=8	SUSC	0.03	SUSC	4	RESIST		
EURL 2018 S-13.6	>64	RESIST	32	RESIST	0.12	NA	0.5	SUSC	0.5	<8	8	SUSC	0.5	SUSC	0.5	<8	<=8	SUSC	8	RESIST	<=1	SUSC	0.5	RESIST
EURL 2018 S-13.7	>64	RESIST	16	SUSC	>32	NA	>64	RESIST	0.5	>=8	16	RESIST	>128	RESIST	2	>=8	>128	RESIST	0.03	SUSC	<=1	SUSC	0.06	SUSC
EURL 2018 S-13.8	>64	RESIST	>64	RESIST	>32	NA	>64	RESIST	0.12	>=8	4	SUSC	16	RESIST	0.5	>=8	<=8	SUSC	0.03	SUSC	<=1	SUSC	<=0.015	SUSC

	Gentamicin GEN		IMIPENEM IMI		MEROPENEM MER		Nalidixic acid NAL		Sulfamethoxazole SMX		TEMOCILLIN TRM		Tetracycline TETRA		TIGECYCLINE TGC		Trimethoprim TMP		ESBL-category	Relevant genes
EURL 2018 S-13.1	<=0.5	SUSC	0.25	SUSC	<=0.03	SUSC	<=4	SUSC	>1024	RESIST	8	SUSC	>64	RESIST	0.5	SUSC	<=0.25	SUSC	ESBL-phenotype	CTX-M-8
EURL 2018 S-13.2	<=0.5	SUSC			<=0.03	SUSC	<=4	SUSC	32	SUSC			<=2	SUSC	<=0.25	SUSC	<=0.25	SUSC	none	ingen
EURL 2018 S-13.3	<=0.5	SUSC			<=0.03	SUSC	<=4	SUSC	32	SUSC			<=2	SUSC	<=0.25	SUSC	<=0.25	SUSC	none	ingen
EURL 2018 S-13.4	1	SUSC	0.25	SUSC	<=0.03	SUSC	>128	RESIST	32	SUSC	8	SUSC	64	RESIST	<=0.25	SUSC	<=0.25	SUSC	ESBL-phenotype	CTX-M-9 TEM-1/1B
EURL 2018 S-13.5	<=0.5	SUSC			<=0.03	SUSC	<=4	SUSC	>1024	RESIST			<=2	SUSC	<=0.25	SUSC	>32	RESIST	none	ingen
EURL 2018 S-13.6	<=0.5	SUSC	2	RESIST	0.5	RESIST	>128	RESIST	<=8	SUSC	>128	RESIST	<=2	SUSC	0.5	SUSC	<=0.25	SUSC	carbapenemase-phenotype	OXA-48
EURL 2018 S-13.7	>32	RESIST	0.5	SUSC	0.06	SUSC	<=4	SUSC	>1024	RESIST	64	RESIST	>64	RESIST	0.5	SUSC	>32	RESIST	ESBL-phenotype	SHV-12 CTX-M-15
EURL 2018 S-13.8	>32	RESIST	0.25	SUSC	<=0.03	SUSC	<=4	SUSC	>1024	RESIST	8	SUSC	<=2	SUSC	0.5	SUSC	>32	RESIST	ESBL-phenotype	CTX-M-3 TEM-1/1B

Resistant

Reference values (MIC-value and interpretation) - *Campylobacter*

Species	Code	Ciprofloxacin CIP		Erythromycin ERY		Gentamicin GEN		Nalidixic acid NAL		Streptomycin STR		Tetracycline TET	
C. jejuni	EURL 2018 C-13.1	4	RESIST	<=1	SUSC	0.25	SUSC	64	RESIST	<=0.25	SUSC	32	RESIST
C. coli	EURL 2018 C-13.2	0.25	SUSC	>128	RESIST	1	SUSC	8	SUSC	2	SUSC	2	SUSC
C. coli	EURL 2018 C-13.3	>16	RESIST	4	SUSC	1	SUSC	>64	RESIST	>16	RESIST	>64	RESIST
C. jejuni	EURL 2018 C-13.4	16	RESIST	>128	RESIST	>16	RESIST	64	RESIST	>16	RESIST	64	RESIST
C. jejuni	EURL 2018 C-13.5	<=0.12	SUSC	<=1	SUSC	0.5	SUSC	4	SUSC	1	SUSC	<=0.5	SUSC
C. jejuni	EURL 2018 C-13.6	8	RESIST	<=1	SUSC	0.25	SUSC	<=1	SUSC	1	SUSC	<=0.5	SUSC
C. coli	EURL 2018 C-13.7	<=0.12	SUSC	>128	RESIST	1	SUSC	8	SUSC	>16	RESIST	<=0.5	SUSC
C. jejuni	EURL 2018 C-13.8	8	RESIST	1	SUSC	0.5	SUSC	>64	RESIST	>16	RESIST	<=0.5	SUSC

Resistant

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G00-06-001/23.06.2017

EURL-AR External Quality Assurance System 2018

- *Salmonella, Campylobacter* and optional genotypic characterisation

Id: «Lab_no_»
«Name»
«Institute__»
«Country»

Kgs. Lyngby, October 2018

Dear «Name»,

Please find enclosed the bacterial strains for the EURL-AR EQAS 2018: eight *Salmonella spp.* and eight *Campylobacter* spp. Upon arrival to your laboratory, the strains should be stored in a dark place at 4°C for stabs, and in a dark and cool place for freeze-dried strains. Charcoal swabs must be subcultured upon arrival.

On the EURL-AR-website (www.eurl-ar.eu) the following documents relevant for this EURL-AR EQAS are available:

- Protocol for antimicrobial susceptibility testing of *Salmonella* and *Campylobacter* and test forms for reporting results
- Instructions for Opening and Reviving Lyophilised Cultures
- Subculture and Maintenance of Quality Control Strains

We ask you to test these *Salmonella* and *Campylobacter* strains for antimicrobial susceptibility. Detailed description of the procedures to follow for antimicrobial susceptibility testing, for optional genotypic characterization and for entering your results into the interactive web database can be found in the protocol. For accessing the database, you need this username and password.

Your username: «Username»

Your password: «Password»

Please keep this document
Your username and password will not appear in other documents

Results should be submitted to the database no later than 7th **December 2018**.

Please acknowledge receipt of this parcel immediately upon arrival (to suska@food.dtu.dk).
Do not hesitate to contact me for further information.

Yours sincerely,

Susanne Karlsmose Pedersen
EURL-AR EQAS-Coordinator

Technical University of Denmark, National Food Institute, Kemitorvet, Building 204, DK-2800 Kgs. Lyngby, Denmark
Ph: +45 3588 6601, e-mail: suska@food.dtu.dk

EU Reference Laboratory for Antimicrobial Resistance
External Quality Assurance System (EQAS) 2018

DTU Food
National Food Institute

PROTOCOL

For antimicrobial susceptibility testing of *Salmonella, Campylobacter* and optional genotypic characterisation of AmpC-, ESBL- and carbapenemase-producing test strains

1 INTRODUCTION

The organisation and implementation of an External Quality Assurance System (EQAS) on antimicrobial susceptibility testing (AST) of *Salmonella* and *Campylobacter* is among the tasks of the EU Reference Laboratory for Antimicrobial Resistance (EURL-AR). The *Salmonella*/*Campylobacter* EQAS 2018 will include AST of eight *Salmonella* and *Campylobacter* strains and AST of reference strains *E. coli* ATCC 25922 (CCM 3954) and *C. jejuni* ATCC 33560 (CCM 6214).

The reference strains are included in the parcel only for new participants of the EQAS who did not receive them previously. The reference strains are original CERTIFIED cultures provided free of charge, and should be used for future internal quality control for antimicrobial susceptibility testing in your laboratory. The reference strains will not be included in the years to come. Therefore, please

take proper care of these strains. Handle and maintain them as suggested in the manual 'Subculture and Maintenance of QC Strains' available on the EURL-AR website (see www.eurl-ar.eu).

Various aspects of the proficiency test scheme may from time to time be subcontracted. When subcontracting occurs it is placed with a competent subcontractor and the National Food Institute is responsible to the scheme participants for the subcontractor's work.

2 OBJECTIVES

This EQAS aims to support laboratories to assess and, if necessary, to improve the quality of results obtained by AST of pathogens of food- and animal-origin, with special regard to *Salmonella* and *Campylobacter*. Further objectives are to evaluate and improve the comparability of surveillance data on antimicrobial susceptibility of *Salmonella* and *Campylobacter* reported to EFSA by different laboratories.

3 OUTLINE OF THE SALM/CAMP EQAS 2018

3.1 Shipping, receipt and storage of strains

In October 2018, the National Reference Laboratories for Antimicrobial Resistance (NRL-AR) will receive a parcel containing eight *Salmonella* and *Campylobacter* strains from the National Food Institute. This parcel will also contain reference strains, but only for participants who did not receive them previously.

All strains belong to UN3373, Biological substance, category B. Extended spectrum beta-lactamase (ESBL)-producing strains as well as carbapenemase producing strains are included in the selected material and are part of the optional EQAS-item, consisting of characterization of genes conferring ESBL- or carbapenemase production. It is the recipients' responsibility to comply with national legislation, rules and regulation regarding the correct use and handling of the provided strains and to possess the proper equipment and protocols to handle these strains.

The reference strains are shipped lyophilised, the *Campylobacter* test strains are shipped as a charcoal swabs and the *Salmonella* test strains are stab cultures. On arrival, the stab cultures and the charcoal swabs must be subcultured, and all cultures should be adequately stored until testing. A suggested procedure for reconstitution of the lyophilised reference strains is presented below.

3.2 QC reference strains

For a suggested procedure for reconstitution of the lyophilised, please refer to the document 'Instructions for opening and reviving lyophilised cultures' on the EURL-AR-website (see www.eurl-ar.eu).

Note that, for the testing of the *E. coli* ATCC25922 reference strain, the two compounds, sulfamethoxazole and sulfisoxazole, are regarded as comparable, i.e. the obtained MIC-value from

the testing of sulfamethoxazole will be evaluated against the acceptance range listed in CLSI M100 for sulfisoxazole.

3.3 Antimicrobial susceptibility testing

The strains should be tested for susceptibility to the antimicrobials listed in Tables 1, 2 and 3, using the method implemented in your laboratory for performing monitoring for EFSA and applying the interpretative criteria listed below.

Participants should perform minimum inhibitory concentration (MIC) determination using the methods stated in the EC regulation EC 652/2013. For interpretation of the results, use the cut-off values listed in Tables 1, 2 and 3. Except where indicated, these represent the current epidemiological cut-off values developed by EUCAST (www.eucast.org), and allow categorisation of bacterial isolates into two categories; resistant or susceptible. A categorisation as intermediate is not accepted.

As the current regulation and recommendations focus on MIC testing only, results obtained by disk diffusion cannot be submitted.

3.3.1 *Salmonella*

The interpretative criteria that should be applied for categorizing the *Salmonella* test strain as resistant or susceptible are those listed in Tables 1 and 2.

Table 1: Antimicrobials recommended for AST of *Salmonella* spp. and interpretative criteria according to table 1 in EC regulation 652/2013

Antimicrobial	MIC (µg/mL) (R>)
Ampicillin (AMP)	8
Azithromycin (AZI)	16*
Cefotaxime (FOT)	0.5
Ceftazidime (TAZ)	2
Chloramphenicol (CHL)	16
Ciprofloxacin (CIP)	0.064
Colistin (COL)	2
Gentamicin (GEN)	2
Meropenem (MERO)	0.125
Nalidixic acid (NAL)	16
Sulfonamides (SMX)	256**
Tetracycline (TET)	8
Tigecycline (TGC)	1***
Trimethoprim (TMP)	2

* Tentative value
** CLSI M100 Table 2A
*** Data from EUCAST is available for *S.* Enteritidis, *S.* Typhimurium, *S.* Typhi and *S.* Paratyphi (for the purpose of this proficiency test, the ECOFF at 1 is applied)

Table 2: Antimicrobials recommended for additional AST of *Salmonella* spp. resistant to cefotaxime, ceftazidime or meropenem and interpretative criteria according to table 4 in EC regulation 652/2013

Antimicrobial	MIC (µg/mL) (R>)
Cefepime, FEP	Not available*
Cefotaxime, FOT	0.5
Cefotaxime + clavulanic acid (F/C)	Not applicable
Cefoxitin, FOX	8
Ceftazidime, TAZ	2
Ceftazidime+ clavulanic acid (T/C)	Not applicable
Ertapenem, ETP	0.06
Imipenem, IMI	1
Meropenem, MERO	0.125
Temocillin, TRM	32**

* Participants are requested to upload the MIC value obtained without selecting an interpretation
** Tentative value

Plasmid-mediated quinolone resistance

When performing antimicrobial susceptibility testing of the *Salmonella* test strains, the interpretative criteria listed in Table 1 for results obtained by MIC-determination should allow detection of plasmid-mediated quinolone resistant test strains.

Beta-lactam- and carbapenem resistance

Confirmatory tests for ESBL production are mandatory on all strains resistant to cefotaxime (FOT), ceftazidime (TAZ) and/or meropenem and should be performed by testing the second panel of antimicrobials (Table 2 in this document corresponding to Table 4 in Commission Implementing Decision 2013/652/EU).

Confirmatory test for AmpC-, ESBL- and carbapenemase production requires use of both cefotaxime (FOT) and ceftazidime (TAZ) alone and in combination with a β-lactamase inhibitor (clavulanic acid). Synergy is defined either as i) a ≥ 3 twofold concentration decrease in an MIC for either antimicrobial agent tested in combination with clavulanic acid vs. the MIC of the agent when tested alone (MIC FOT:FOT/Cl or TAZ:TAZ/Cl ratio ≥ 8) (CLSI M100 Table 3A, Tests for ESBLs). The presence of synergy indicates ESBL production.

Confirmatory test for carbapenemase production requires the testing of meropenem (MERO).

Detection of AmpC-type beta-lactamases can be performed by testing the bacterium for susceptibility to cefoxitin (FOX). Resistance to FOX could indicate the presence of an AmpC-type beta-lactamase.

The classification of the phenotypic results should be based on the most recent EFSA recommendations (available in The European Union summary report on antimicrobial resistance in

zoonotic and indicator bacteria from humans, animals and food in 2015, EFSA Journal 2017;15(2):4694,212 pp. (page 43), and in the appendix to this protocol). It is important to notice that two cut-off values apply for cefotaxime and ceftazidime: the EUCAST cut-off values (ECOFFs: FOT>0.5 and TAZ>2), which are those used to define R/S, and the screening cut-off values (FOT>1 and TAZ>1), which are those applied to categorise bacterial phenotypes as ESBL, AmpC, carbapenemase, etc. based on panel 2 results (see Appendix).

3.3.2 *Campylobacter*

The obtained values of the *C. jejuni* QC reference strain will be evaluated according to the values listed in the CLSI document VET06, 1st ed., i.e. based on incubation at 36-37ºC for 48 hours or 42ºC for 24 hours.

Table 3: Antimicrobials recommended for AST of *Campylobacter jejuni* and *C. coli* and interpretative criteria according to table 1 in EC regulation 652/2013

Antimicrobial	*C. jejuni*	*C. coli*
	MIC (µg/mL) (R>)	MIC (µg/mL) (R>)
Ciprofloxacin (CIP)	0.5	0.5
Erythromycin (ERY)	4	8
Gentamicin (GEN)	2	2
Nalidixic acid (NAL)	16	16
Streptomycin (STR)	4	4
Tetracycline (TET)	1	2

Identification of *Campylobacter* species

Species identification of the *Campylobacter* test strains must be performed by the NRLs using in-house methods or adopting the protocol available on the EURL-AR website under: http://eurl-ar.eu/233-protocols.htm.

3.4 Optional genotypic characterisation

For the optional genotypic characterisation of the AmpC-, ESBL- or carbepenemase producing *Salmonella* test strains, the requested results are the genes encoding AmpC-, ESBL- or carbepenemase –production. AmpC-, ESBL- or carbapenemase types included in the test are the following: ACC, ACT, CARB, CMY, CTX-M, DHA, FOX, GES, IMP, KPC, MOX, NDM, OXA, PER, SCO, SHV, TEM, VEB, and VIM. The database lists the relevant variants of each type.

When uploading the results in the database, the identified genes will be evaluated against the expected results. The results will be evaluated on the detected type (ACC-, ACT-, CARB-, etc.) as well as the variant identified.

The method used for the genotypic characterisation should be your laboratory's routine method. The expected results listed in the database are those obtained by the EURL-AR.

4 REPORTING OF RESULTS AND EVALUATION

Test forms are available for recording your results before you enter them into the interactive web database.

We recommend reading carefully the description reported in paragraph 5 before entering your results in the web database. **Results must be submitted no later than December 7th 2018.** After the deadline when all participants have uploaded results, you will be able to login to the database once again, and to view and print an automatically generated report evaluating your results. Results in agreement with the expected interpretation are categorised as 'correct', while results deviating from the expected interpretation are categorised as 'incorrect'.

If you experience difficulties in entering your results, please contact us directly.

All results will be summarized in a report which will be publicly available. The data in the report will be presented with laboratory codes. A laboratory code is known to the individual laboratory, whereas the complete list of laboratories and their codes is confidential and known only to the EURL-AR and the EU Commission. All conclusions will be public.

If you have questions, please do not hesitate to contact the EQAS Coordinator:

Susanne Karlsmose Pedersen
National Food Institute,
Technical University of Denmark
Kemitorvet, Building 204, DK-2800 Lyngby
Denmark
Tel: +45 3588 6601
E-mail: suska@food.dtu.dk

5 HOW TO ENTER RESULTS IN THE INTERACTIVE DATABASE

Please read carefully this paragraph before entering the web page.

Remember that you need by your side the completed test forms.

Enter the EURL-AR EQAS start web page (http://eurl-ar.food.dtu.dk), write your username and password (lower-case) and press enter. Your username and password are indicated in the letter following your strains. Do not hesitate to contact us if you experience problems with the login.

You can browse back and forth by using the Home or back keys, but please remember to save your inputs before.

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Click on either "*Salmonella* test results" or "*Campylobacter* test results" for input of test results.

Click on "Start of Data Entry - Methods"

In the next page, you navigate among fields with the Tab-key and the mouse.

Complete the fields related to the method used for antimicrobial susceptibility testing and the brand of MIC trays, etc.

When submitting *Campylobacter* results, fill in the incubation conditions applied for susceptibility testing of *Campylobacter* – 36°C/48h or 42°C/24h.

Click on "save and go to next page"

In the data entry pages, you enter the species (for *Campylobacter* only), the obtained MIC-value and the interpretation (R, resistant or S, susceptible) for each *Salmonella* and *Campylobacter* strain.

For *Salmonella*, remember to also report the results for the ESBL detection tests.

If you did not test for susceptibility to a given antimicrobial, please leave the field empty.

Click on "save and go to next page"

When uploading data on the reference strains, please enter MIC values in µg/ml. Remember to use the operator keys to show symbols like "equal to", etc.

Click on "save".

Review the input pages by browsing through them and make corrections if necessary. Remember to save a page if you make corrections. If you press home a page without saving changes, you will see an error screen. In this case, click on "save" to save your results, browse back to the page and then continue.

Please complete the evaluation form.

Before approving your input, please be sure that you have filled in all the relevant fields as YOU CAN ONLY APPROVE ONCE! The approval blocks your data entry in the interactive database.

If you have performed the optional genotypic characterisation:

Click on "Gene test" and follow the description in the database for upload of the results of the optional genotypic characterization. Approve your input. Be sure that you have filled in all the results before approval. The approval blocks your data entry in the interactive database, but allows you to see the submitted results.

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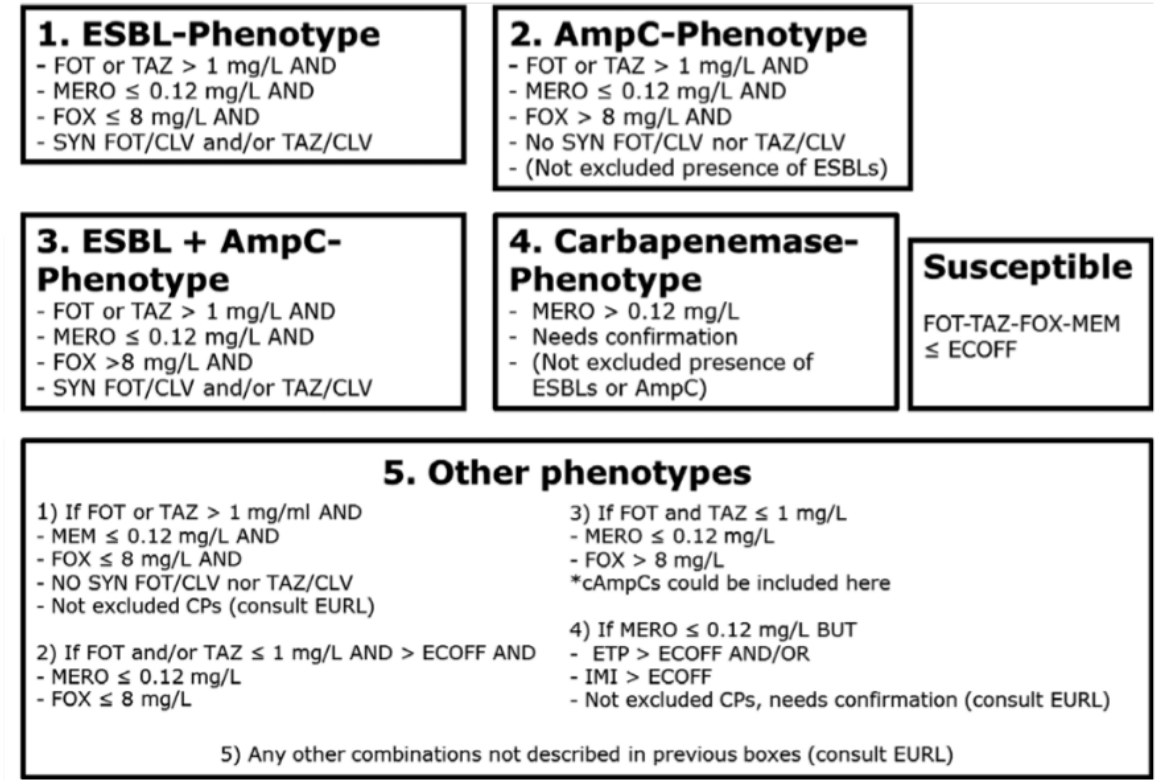
APPENDIX

Criteria for interpretation of *Salmonella*, panel 2 results



Please refer to: EFSA (European Food Safety Authority) and ECDC (European Centre for Disease Prevention and Control), 2017. The European Union summary report on antimicrobial resistance in zoonotic and indicator bacteria from humans, animals and food in 2015. EFSA Journal 2017;15(2):4694, 212 pp. doi:10.2903/j.efsa.2017.4694 (page 43).

Salmonella, Campylobacter and genetic characterisation

TEST FORMS

Name:
Name of laboratory:
Name of institute:
City:
Country:
E-mail:
Fax:

Comments:

TEST FORM

Does your laboratory have an accreditation for performing *Salmonella* AST? ☐ Yes ☐ No

Which method did you use for antimicrobial susceptibility testing of *Salmonella* in this EQAS:

☐ Broth microdilution

Brand of microbroth plates/agar**:**
Incubation conditions: °C/ h

How many *Salmonella* isolates does your laboratory annually isolate:

How many *Salmonella* isolates does your laboratory annually test for antimicrobial susceptibility by a MIC method:

Which method was followed for the preparation of the inoculum (please describe)

- Which standard was followed (TREK, CLSI…)
- Which solvent was used for the preparation of the 0.5 McFarland solution (water, saline)
- Please describe in detail how you prepared the dilution of the inoculum (including the volume in final MH-dilution and intended dilution level; e.g. diluted 1:1000 by adding 10µl of 0.5 McFarland solution in 10ml MH broth, for an expected inoculum of $1*10^5$ CFU/ml)

Comments or additional information:

TEST FORM

Does your laboratory have an accreditation for *Campylobacter* AST? ☐ Yes ☐ No

Incubation conditions: ☐ 36-37ºC / 48h ☐ 42ºC / 24h

Method used for antimicrobial susceptibility testing of *Campylobacter* in this EQAS:
☐ Broth microdilution

Brand of microbroth plates/agar:

How many *Campylobacter* isolates does your laboratory annually isolate:

How many *Campylobacter* isolates does your laboratory annually susceptibility test:

Which method was followed for the preparation of the inoculum (please describe)

- Which standard was followed (TREK, CLSI…)
- Which solvent was used for the preparation of the 0.5 McFarland solution (water, saline)
- Please describe in detail how you prepared the dilution of the inoculum (including the volume in final MH-dilution and intended dilution level; e.g. diluted 1:1000 by adding 10µl of 0.5 McFarland solution in 10ml MH broth, for an expected inoculum of $1*10^5$ CFU/ml)

Comments or additional information:

TEST FORM

Strain	Antimicrobial	Results and interpretation		
		≤ / >	MIC-value (µg/ml)	S / R
Salmonella EURL S. 13.X	Ampicillin, AMP			
	Azithromycin, AZI			
	Cefotaxime, FOT			
	Ceftazidime, TAZ			
	Chloramphenicol, CHL			
	Ciprofloxacin CIP			
	Colistin, COL			
	Gentamicin, GEN			
	Meropenem, MERO			
	Nalidixic acid, NAL			
	Sulfamethoxazole, SMX			
	Tetracycline, TET			
	Tigecycline, TGC			
	Trimethoprim, TMP			

All strains resistant to cefotaxime (FOT), ceftazidime (TAZ) or meropenem (MERO) must be included for testing in the second panel as part of confirmatory tests for ESBL-, AmpC or carbapenemase production. See further description in the protocol section '3.3.1 *Salmonella*'.

Strain	Antimicrobial	Results and interpretation		
		≤ / >	MIC-value (µg/ml)	S / R
Salmonella EURL S. 13.X	Cefepime, FEP			
	Cefotaxime, FOT			
	Cefotaxime + clavulanic acid (F/C)			
	Cefoxitin, FOX			
	Ceftazidime, TAZ			
	Ceftazidime+ clavulanic acid (T/C)			
	Ertapenem, ETP			
	Imipenem, IMI			
	Meropenem, MERO			
	Temocillin, TRM			

Interpretation of PANEL 2 results:

☐ Presumptive ESBL ☐ Presumptive AmpC ☐ Other phenotype
☐ Presumptive ESBL+ AmpC ☐ Presumptive Carbapenemase ☐ Susceptible

Comments (include optional genotype or other results):

TEST FORM

Antimicrobial susceptibility testing of reference strain *E. coli* ATCC 25922

	Antimicrobial	MIC-value (µg/ml)
1st panel	Ampicillin, AMP	
	Azithromycin, AZI	
	Cefotaxime, FOT	
	Ceftazidime, TAZ	
	Chloramphenicol, CHL	
	Ciprofloxacin, CIP	
	Colistin, COL	
	Gentamicin, GEN	
	Meropenem, MERO	
	Nalidixic acid, NAL	
	Sulfamethoxazole, SMX*	
	Tetracycline, TET	
	Tigecycline, TGC	
	Trimethoprim, TMP	
2nd panel	Cefepime, FEP	
	Cefotaxime, FOT	
	Cefotaxime + clavulanic acid (F/C)	
	Cefoxitin, FOX	
	Ceftazidime, TAZ	
	Ceftazidime+ clavulanic acid (T/C)	
	Ertapenem, ETP	
	Imipenem, IMI	
	Meropenem, MERO	
	Temocillin, TRM	

* for the testing of the *E. coli* ATCC25922 reference strain, sulfamethoxazole and sulfisoxazole, are regarded as comparable, i.e. the obtained MIC-value from the testing of sulfamethoxazole will be evaluated against the acceptance range listed in CLSI M100 for sulfisoxazole (CLSI M100, Table 3).

TEST FORM

Strain	Antimicrobial	Interpretation	
		MIC-value (µg/ml)	S / R
Campylobacter EURL C-13.X ☐ *C. jejuni* ☐ *C. coli*	Ciprofloxacin		
	Erythromycin		
	Gentamicin		
	Nalidixic acid		
	Streptomycin		
	Tetracycline		
Campylobacter EURL C-13.X ☐ *C. jejuni* ☐ *C. coli*	Ciprofloxacin		
	Erythromycin		
	Gentamicin		
	Nalidixic acid		
	Streptomycin		
	Tetracycline		
Campylobacter EURL C-13.X ☐ *C. jejuni* ☐ *C. coli*	Ciprofloxacin		
	Erythromycin		
	Gentamicin		
	Nalidixic acid		
	Streptomycin		
	Tetracycline		
Campylobacter EURL C-13.X ☐ *C. jejuni* ☐ *C. coli*	Ciprofloxacin		
	Erythromycin		
	Gentamicin		
	Nalidixic acid		
	Streptomycin		
	Tetracycline		

TEST FORM

Susceptibility testing of *Campylobacter jejuni* reference strain ATCC 33560

Strain	Antimicrobial	MIC-value (µg/ml)	
		36 °C/48 hours	42 °C/24 hours
C. jejuni ATCC 33560	Ciprofloxacin		
	Erythromycin		
	Nalidixic acid		
	Tetracycline		

For Agar dilution:

Susceptibility testing of *Campylobacter jejuni* reference strain ATCC 33560

Strain	Antimicrobial	MIC-value (µg/ml)
C. jejuni ATCC 33560	Ciprofloxacin	
	Erythromycin	
	Gentamicin	
	Nalidixic acid	
	Tetracycline	

TEST FORM – genotypic characterisation

Genotypic characterisation of the test strains

Strain code:	Method used: If PCR-methods, additional information should be given below
Gene: ☐ Found ☐ Tested, not found	☐ Published method , reference:
	☐ In-house method
	Primer used 5'→3':
	Primer used 3'→5':
Gene: ☐ Found ☐ Tested, not found	☐ Published method , reference:
	☐ In-house method
	Primer used 5'→3':
	Primer used 3'→5':
Gene: ☐ Found ☐ Tested, not found	☐ Published method , reference:
	☐ In-house method
	Primer used 5'→3':
	Primer used 3'→5':
Gene: ☐ Found ☐ Tested, not found	☐ Published method , reference:
	☐ In-house method
	Primer used 5'→3':
	Primer used 3'→5':
Gene: ☐ Found ☐ Tested, not found	☐ Published method , reference:
	☐ In-house method
	Primer used 5'→3':
	Primer used 3'→5':

Comments:

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INSTRUCTIONS FOR OPENING AND REVIVING LYOPHILISED CULTURES

Instructions adjusted from Czech Collection of Microorganisms (CCM) document 'Instructions for Opening and Reviving of Freeze-Dried Bacteria and Fungi' available on http://www.sci.muni.cz.

Lyophilised cultures are supplied in vacuum-sealed ampoules. Care should be taken in opening the ampoule. All instructions given below should be followed closely to ensure the safety of the person who opens the ampoule and to prevent contamination of the culture.

a. Check the number of the culture on the label inside the ampoule
b. Make a file cut on the ampoule near the middle of the plug (see Figure 1)
c. Disinfect the ampoule with alcohol-dampened gauze or alcohol-dampened cotton wool from just below the plug to the pointed end
d. Apply a red-hot glass rod to the file cut to crack the glass and allow air to enter slowly into the ampoule
e. Remove the pointed end of the ampoule into disinfectant
f. Add about 0.3 ml appropriate broth to the dried suspension using a sterile Pasteur pipette and mix carefully to avoid creating aerosols. Transfer the contents to one or more suitable solid and /or liquid media
g. Incubate the inoculated medium at appropriate conditions for several days
h. Autoclave or disinfect effectively the used Pasteur pipette, the plug and all the remains of the original ampoule before discarding

Notes:

- Cultures should be grown on media and under conditions as recommended in the CCM catalogue (see http://www.sci.muni.cz)
- Cultures may need at least one subculturing before they can be optimally used in experiments
- Unopened ampoules should be kept in a dark and cool place!

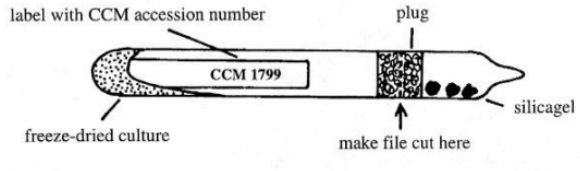

Figure 1: from CCM document 'Instructions for Opening and Reviving of Freeze-Dried Bacteria and Fungi' available on http://www.sci.muni.cz

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SUBCULTURE AND MAINTENANCE OF QUALITY CONTROL STRAINS

1 PURPOSE AND REFERENCES

Improper storage and repeated subculturing of bacteria can produce alterations in antimicrobial susceptibility test results. The Clinical and Laboratory Standards Institute (CLSI) has published guidelines for Quality Control (QC) stock culture maintenance to ensure consistent antimicrobial susceptibility test (AST) results.

The following can be regarded as a summary of information that should be followed for subculturing and maintaining QC-strains when performing AST by broth dilution methods. For full information related to this subject, the following standards are relevant: M100 (Performance Standards for Antimicrobial Susceptibility Testing) and M7 (Methods for Dilution Antimicrobial Susceptibility Test for Bacteria That Grow Aerobically; Approved Standard).

2 DEFINITION OF TERMS

Reference Culture: A reference culture is a microorganism preparation that is acquired from a culture type collection.

Reference Stock Culture: A reference stock culture is a microorganism preparation that is derived from a reference culture. Guidelines and standards outline how reference stock cultures must be processed and stored.

Working Stock Cultures: A working stock culture is growth derived from a reference stock culture. Guidelines and standards outline how working stock cultures must be processed and how often they can be subcultured.

Subcultures (Passages): A subculture is simply the transfer of established microorganism growth on media to fresh media. The subsequent growth on the fresh media constitutes a subculture or passage. Growing a reference culture or reference stock culture from its preserved status (frozen or lyophilized) is not a subculture. The preserved microorganism is not in a stage of established growth until it is thawed or hydrated and grown for the first time.

3 IMPORTANT CONSIDERATIONS

- Do not use disc diffusion strains for MIC determination.
- Obtain QC strains from a reliable source such as ATCC.
- CLSI requires that QC be performed either on the same day or weekly (after QC-validation).
- Any changes in materials or procedure must be validated with QC before implemented
- For example: Agar and broth methods may give different QC ranges for drugs such as glycopeptides, aminoglycosides and macrolides.

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- Periodically perform colony counts to check the inoculum preparation procedure.
- Ideally, test values should be in the middle of the acceptable range.
- Graphing QC data points over time can help identify changes in data helpful for troubleshooting problems.

4 STORAGE OF REFERENCE STRAINS

Preparation of stock cultures

- Use a suitable stabilizer such as 50% fetal calf serum in broth, 10-15% glycerol in tryptic soy broth, defibrinated sheep blood or skim milk to prepare multiple aliquots.
- Store at -20°C, -70°C or liquid nitrogen (alternatively, freeze dry.)
- Before using rejuvenated strains for QC, subculture to check for purity and viability.

Working cultures

- Set up on agar slants with appropriate medium, store at 4-8°C and subculture weekly.
- Replace the working strain with a stock culture at least monthly.
- If a change in the organisms inherent susceptibility occurs, obtain a fresh stock culture or a new strain from a reference culture collection e.g. ATCC.

5 FREQUENCY OF TESTING

Weekly vs. daily testing

Weekly testing is possible if the laboratory can demonstrate satisfactory performance with daily testing according to the descriptions in the CLSI guidelines.

- Documentation showing reference strain results from 20 or 30 consecutive test days were within the acceptable range.
- For each antimicrobial/organism combination, no more one out of 20 or three out of 30 MIC values may be outside the acceptable range.

When the above are fulfilled, each quality control strain may be tested once a week and whenever any reagent component is changed.

Corrective Actions

If an MIC is outside the range in weekly testing, corrective action is required as follows:

- Repeat the test if there is an obvious error e.g. wrong strain or incubation conditions used
- If there is no obvious error, return to daily control testing

If five acceptable QC results are available, no additional days of QC-testing are needed.

If the problem cannot be resolved, continue daily testing until the errors are identified.

Repeat the 30 days validation before resuming weekly testing.

Quality Control ranges for ATCC reference strains

E. coli ATCC 25922	
Antimicrobial	**MIC**
Ampicillin, AMP	2-8
Azithromycin, AZI	none
Cefepime, FEP	0.016-0.12
Cefotaxime, FOT	0.03-0.12
Cefotaxime + clavulanic acid, F/C	none
Cefoxitin, FOX	2-8
Ceftazidime, TAZ	0.06-0.5
Ceftazidime + clavulanic acid, T/C	none
Chloramphenicol, CHL	2-8
Ciprofloxacin, CIP	0.004-0.016
Colistin, COL	0.25-2
Ertapenem, ETP	0.004-0.016
Gentamicin, GEN	0.25-1
Imipenem, IMI	0.06-0.25
Meropenem, MERO	0.008-0.06
Nalidixic acid, NAL	1-4
Sulfamethoxazole, SMX	8-32
Temocillin, TRM	none
Tetracycline, TET	0.5-2
Tigecycline, TGC	0.03-0.25
Trimethoprim, TMP	0.5-2

MIC ranges and disc diffusion ranges are according to CLSI M100 28th edition

Campylobacter jejuni ATCC 33560				
Antimicrobial	**Microbroth (36-37°C/48h)**	**Microbroth (42°C/24h)**	**Agar dilution (36-37°C/48h)**	**Agar dilution (42°C/24h)**
Ciprofloxacin, CIP	0.06-0.25	0.03-0.12	0.12-1	0.06-0.5
Erythromycin, ERY	0.5-2	0.25-2	1-8	1-4
Gentamicin, GEN	0.5-2	0.25-2	0.5-2	0.5-4
Nalidixic acid, NAL	4-16	4-16	None	None
Tetracycline, TET	0.25-2	0.25-1	None	None

Ranges are according to CLSI (VET06, 1st ed.)

Test results from the reference strain *E. coli* ATCC 25922 obtained by microbroth dilution

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
2	1	Ampicillin	=	8	2	8	1	35±1	18-24
2	1	Cefotaxime	<=	0.25	0.03	0.12	1	35±1	18-24
2	1	Ceftazidime	<=	0.5	0.06	0.5	1	35±1	18-24
2	1	Chloramphenicol	<=	8	2	8	1	35±1	18-24
2	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35±1	18-24
2	1	Colistin	<=	1	0.25	2	1	35±1	18-24
2	1	Gentamicin	<=	0.5	0.25	1	1	35±1	18-24
2	1	Meropenem	<=	0.03	0.008	0.06	1	35±1	18-24
2	1	Nalidixic acid	<=	4	1	4	1	35±1	18-24
2	1	Sulfamethoxazole	=	32	8	32	1	35±1	18-24
2	1	Tetracycline	<=	2	0.5	2	1	35±1	18-24
2	1	Tigecycline	<=	0.25	0.03	0.25	1	35±1	18-24
2	1	Trimethoprim	=	0.5	0.5	2	1	35±1	18-24
2	2	Cefepime	<=	0.06	0.016	0.12	1	35±1	18-24
2	2	Cefotaxime	<=	0.25	0.03	0.12	1	35±1	18-24
2	2	Cefoxitin	=	8	2	8	1	35±1	18-24
2	2	Ceftazidime	=	0.5	0.06	0.5	1	35±1	18-24
2	2	Ertapenem	<=	0.015	0.004	0.016	1	35±1	18-24
2	2	Imipenem	=	0.25	0.06	0.25	1	35±1	18-24
2	2	Meropenem	<=	0.03	0.008	0.06	1	35±1	18-24
4	1	Ampicillin	=	4	2	8	1	37°C	24
4	1	Cefotaxime	<=	0.25	0.03	0.12	1	37°C	24
4	1	Ceftazidime	<=	0.5	0.06	0.5	1	37°C	24
4	1	Chloramphenicol	<=	8	2	8	1	37°C	24
4	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37°C	24
4	1	Colistin	<=	1	0.25	2	1	37°C	24
4	1	Gentamicin	<=	**0.05**	0.25	1	**0**	37°C	24
4	1	Meropenem	<=	0.03	0.008	0.06	1	37°C	24
4	1	Nalidixic acid	<=	4	1	4	1	37°C	24
4	1	Sulfamethoxazole	=	32	8	32	1	37°C	24
4	1	Tetracycline	<=	2	0.5	2	1	37°C	24
4	1	Tigecycline	=	**0.5**	0.03	0.25	**0**	37°C	24
4	1	Trimethoprim	=	1	0.5	2	1	37°C	24
4	2	Cefepime	<=	0.06	0.016	0.12	1	37°C	24
4	2	Cefotaxime	<=	0.25	0.03	0.12	1	37°C	24
4	2	Cefoxitin	=	2	2	8	1	37°C	24
4	2	Ceftazidime	=	0.5	0.06	0.5	1	37°C	24
4	2	Ertapenem	<=	0.015	0.004	0.016	1	37°C	24
4	2	Imipenem	<=	0.12	0.06	0.25	1	37°C	24
4	2	Meropenem	<=	0.03	0.008	0.06	1	37°C	24
6	1	Ampicillin	=	4	2	8	1	35	18
6	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
6	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18
6	1	Chloramphenicol	<=	8	2	8	1	35	18
6	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18
6	1	Colistin	<=	1	0.25	2	1	35	18
6	1	Gentamicin	<=	0.5	0.25	1	1	35	18
6	1	Meropenem	<=	0.03	0.008	0.06	1	35	18
6	1	Nalidixic acid	<=	4	1	4	1	35	18
6	1	Sulfamethoxazole	=	32	8	32	1	35	18
6	1	Tetracycline	<=	2	0.5	2	1	35	18
6	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18
6	1	Trimethoprim	=	1	0.5	2	1	35	18
6	2	Cefepime	<=	0.06	0.016	0.12	1	35	18
6	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
6	2	Cefoxitin	=	4	2	8	1	35	18
6	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	18
6	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18
6	2	Imipenem	=	0.25	0.06	0.25	1	35	18
6	2	Meropenem	<=	0.03	0.008	0.06	1	35	18
9	1	Ampicillin	=	4	2	8	1	35+-1	20
9	1	Ceftazidime	<=	0.5	0.06	0.5	1	35+-1	20
9	1	Chloramphenicol	<=	8	2	8	1	35+-1	20
9	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35+-1	20
9	1	Colistin	<=	1	0.25	2	1	35+-1	20
9	1	Gentamicin	<=	0.5	0.25	1	1	35+-1	20
9	1	Meropenem	<=	0.03	0.008	0.06	1	35+-1	20
9	1	Nalidixic acid	<=	4	1	4	1	35+-1	20
9	1	Sulfamethoxazole	=	16	8	32	1	35+-1	20
9	1	Tetracycline	<=	2	0.5	2	1	35+-1	20
9	1	Tigecycline	<=	0.25	0.03	0.25	1	35+-1	20
9	1	Trimethoprim	=	1	0.5	2	1	35+-1	20
9	2	Cefepime	<=	0.06	0.016	0.12	1	35+-1	20
9	2	Cefoxitin	=	4	2	8	1	35+-1	20
9	2	Ceftazidime	<=	0.25	0.06	0.5	1	35+-1	20
9	2	Ertapenem	<=	0.015	0.004	0.016	1	35+-1	20
9	2	Imipenem	<=	0.12	0.06	0.25	1	35+-1	20
9	2	Meropenem	<=	0.03	0.008	0.06	1	35+-1	20

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
11	1	Ampicillin	=	4	2	8	1	37	20
11	1	Cefotaxime	<=	0.25	0.03	0.12	1	37	20
11	1	Ceftazidime	<=	0.5	0.06	0.5	1	37	20
11	1	Chloramphenicol	<=	8	2	8	1	37	20
11	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37	20
11	1	Colistin	<=	1	0.25	2	1	37	20
11	1	Gentamicin	=	1	0.25	1	1	37	20
11	1	Meropenem	<=	0.03	0.008	0.06	1	37	20
11	1	Nalidixic acid	<=	4	1	4	1	37	20
11	1	Sulfamethoxazole	=	16	8	32	1	37	20
11	1	Tetracycline	<=	2	0.5	2	1	37	20
11	1	Tigecycline	<=	0.25	0.03	0.25	1	37	20
11	1	Trimethoprim	=	0.5	0.5	2	1	37	20
11	2	Cefepime	<=	0.06	0.016	0.12	1	37	20
11	2	Cefotaxime	<=	0.25	0.03	0.12	1	37	20
11	2	Cefoxitin	=	4	2	8	1	37	20
11	2	Ceftazidime	<=	0.25	0.06	0.5	1	37	20
11	2	Ertapenem	<=	0.015	0.004	0.016	1	37	20
11	2	Imipenem	<=	0.12	0.06	0.25	1	37	20
11	2	Meropenem	<=	0.03	0.008	0.06	1	37	20
12	1	Ampicillin	=	4	2	8	1	35	18
12	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
12	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18
12	1	Chloramphenicol	<=	8	2	8	1	35	18
12	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18
12	1	Colistin	<=	1	0.25	2	1	35	18
12	1	Gentamicin	<=	0.5	0.25	1	1	35	18
12	1	Meropenem	<=	0.03	0.008	0.06	1	35	18
12	1	Nalidixic acid	<=	4	1	4	1	35	18
12	1	Sulfamethoxazole	=	16	8	32	1	35	18
12	1	Tetracycline	<=	2	0.5	2	1	35	18
12	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18
12	1	Trimethoprim	=	0.5	0.5	2	1	35	18
12	2	Cefepime	<=	0.06	0.016	0.12	1	35	18
12	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
12	2	Cefoxitin	=	4	2	8	1	35	18
12	2	Ceftazidime	=	0.5	0.06	0.5	1	35	18
12	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18
12	2	Imipenem	<=	0.12	0.06	0.25	1	35	18
12	2	Meropenem	<=	0.03	0.008	0.06	1	35	18
16	1	Ampicillin	=	4	2	8	1	35	18-24
16	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
16	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18-24
16	1	Chloramphenicol	<=	8	2	8	1	35	18-24
16	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18-24
16	1	Colistin	<=	1	0.25	2	1	35	18-24
16	1	Gentamicin	<=	0.5	0.25	1	1	35	18-24
16	1	Meropenem	<=	0.03	0.008	0.06	1	35	18-24
16	1	Nalidixic acid	<=	4	1	4	1	35	18-24
16	1	Sulfamethoxazole	=	32	8	32	1	35	18-24
16	1	Tetracycline	<=	2	0.5	2	1	35	18-24
16	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18-24
16	1	Trimethoprim	=	0.5	0.5	2	1	35	18-24
16	2	Cefepime	<=	0.06	0.016	0.12	1	35	18-24
16	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
16	2	Cefoxitin	=	4	2	8	1	35	18-24
16	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	18-24
16	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18-24
16	2	Imipenem	=	0.25	0.06	0.25	1	35	18-24
16	2	Meropenem	<=	0.03	0.008	0.06	1	35	18-24
17	1	Ampicillin	=	8	2	8	1	36	18-20
17	1	Cefotaxime	<=	0.25	0.03	0.12	1	36	18-20
17	1	Ceftazidime	<=	0.5	0.06	0.5	1	36	18-20
17	1	Chloramphenicol	<=	8	2	8	1	36	18-20
17	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	36	18-20
17	1	Colistin	<=	1	0.25	2	1	36	18-20
17	1	Gentamicin	=	1	0.25	1	1	36	18-20
17	1	Meropenem	<=	0.03	0.008	0.06	1	36	18-20
17	1	Nalidixic acid	<=	4	1	4	1	36	18-20
17	1	Sulfamethoxazole	<=	8	8	32	1	36	18-20
17	1	Tetracycline	<=	2	0.5	2	1	36	18-20
17	1	Tigecycline	<=	0.25	0.03	0.25	1	36	18-20
17	1	Trimethoprim	<=	**0.25**	0.5	2	**0**	36	18-20
17	2	Cefepime	<=	0.06	0.016	0.12	1	36	18-20
17	2	Cefotaxime	<=	0.25	0.03	0.12	1	36	18-20
17	2	Cefoxitin	=	4	2	8	1	36	18-20
17	2	Ceftazidime	=	0.5	0.06	0.5	1	36	18-20
17	2	Ertapenem	<=	0.015	0.004	0.016	1	36	18-20
17	2	Imipenem	=	0.25	0.06	0.25	1	36	18-20
17	2	Meropenem	<=	0.03	0.008	0.06	1	36	18-20

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
18	1	Ampicillin	=	2	2	8	1	35	18
18	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
18	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18
18	1	Chloramphenicol	<=	8	2	8	1	35	18
18	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18
18	1	Colistin	<=	1	0.25	2	1	35	18
18	1	Gentamicin	<=	0.5	0.25	1	1	35	18
18	1	Meropenem	<=	0.03	0.008	0.06	1	35	18
18	1	Nalidixic acid	<=	4	1	4	1	35	18
18	1	Sulfamethoxazole	=	16	8	32	1	35	18
18	1	Tetracycline	<=	2	0.5	2	1	35	18
18	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18
18	1	Trimethoprim	=	0.5	0.5	2	1	35	18
18	2	Cefepime	<=	0.06	0.016	0.12	1	35	18
18	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
18	2	Cefoxitin	=	4	2	8	1	35	18
18	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	18
18	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18
18	2	Imipenem	<=	0.12	0.06	0.25	1	35	18
18	2	Meropenem	<=	0.03	0.008	0.06	1	35	18
19	1	Ampicillin	=	4	2	8	1	35	18
19	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
19	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18
19	1	Chloramphenicol	<=	8	2	8	1	35	18
19	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18
19	1	Colistin	<=	1	0.25	2	1	35	18
19	1	Gentamicin	<=	0.5	0.25	1	1	35	18
19	1	Meropenem	<=	0.03	0.008	0.06	1	35	18
19	1	Nalidixic acid	<=	4	1	4	1	35	18
19	1	Sulfamethoxazole	=	16	8	32	1	35	18
19	1	Tetracycline	<=	2	0.5	2	1	35	18
19	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18
19	1	Trimethoprim	=	0.5	0.5	2	1	35	18
19	2	Cefepime	<=	0.06	0.016	0.12	1	35	18
19	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18
19	2	Cefoxitin	=	4	2	8	1	35	18
19	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	18
19	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18
19	2	Imipenem	<=	0.12	0.06	0.25	1	35	18
19	2	Meropenem	<=	0.03	0.008	0.06	1	35	18
20	1	Ampicillin	=	4	2	8	1	37C +/-1C	20h +/-2h
20	1	Cefotaxime	<=	0.25	0.03	0.12	1	37C +/-1C	20h +/-2h
20	1	Ceftazidime	<=	0.5	0.06	0.5	1	37C +/-1C	20h +/-2h
20	1	Chloramphenicol	<=	8	2	8	1	37C +/-1C	20h +/-2h
20	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37C +/-1C	20h +/-2h
20	1	Colistin	<=	1	0.25	2	1	37C +/-1C	20h +/-2h
20	1	Gentamicin	<=	0.5	0.25	1	1	37C +/-1C	20h +/-2h
20	1	Meropenem	<=	0.03	0.008	0.06	1	37C +/-1C	20h +/-2h
20	1	Nalidixic acid	<=	4	1	4	1	37C +/-1C	20h +/-2h
20	1	Sulfamethoxazole	=	16	8	32	1	37C +/-1C	20h +/-2h
20	1	Tetracycline	<=	2	0.5	2	1	37C +/-1C	20h +/-2h
20	1	Tigecycline	<=	0.25	0.03	0.25	1	37C +/-1C	20h +/-2h
20	1	Trimethoprim	=	0.5	0.5	2	1	37C +/-1C	20h +/-2h
20	2	Cefepime	<=	0.06	0.016	0.12	1	37C +/-1C	20h +/-2h
20	2	Cefotaxime	<=	0.25	0.03	0.12	1	37C +/-1C	20h +/-2h
20	2	Cefoxitin	=	4	2	8	1	37C +/-1C	20h +/-2h
20	2	Ceftazidime	<=	0.25	0.06	0.5	1	37C +/-1C	20h +/-2h
20	2	Ertapenem	<=	0.015	0.004	0.016	1	37C +/-1C	20h +/-2h
20	2	Imipenem	=	0.25	0.06	0.25	1	37C +/-1C	20h +/-2h
20	2	Meropenem	<=	0.03	0.008	0.06	1	37C +/-1C	20h +/-2h
21	1	Ampicillin	=	4	2	8	1	36	24
21	1	Cefotaxime	<=	0.25	0.03	0.12	1	36	24
21	1	Ceftazidime	=	0.5	0.06	0.5	1	36	24
21	1	Chloramphenicol	<=	8	2	8	1	36	24
21	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	36	24
21	1	Colistin	<=	1	0.25	2	1	36	24
21	1	Gentamicin	<=	0.5	0.25	1	1	36	24
21	1	Meropenem	<=	0.03	0.008	0.06	1	36	24
21	1	Nalidixic acid	<=	4	1	4	1	36	24
21	1	Sulfamethoxazole	=	16	8	32	1	36	24
21	1	Tetracycline	=	2	0.5	2	1	36	24
21	1	Tigecycline	<=	0.25	0.03	0.25	1	36	24
21	1	Trimethoprim	<=	0.5	0.5	2	1	36	24
21	2	Cefepime	<=	0.06	0.016	0.12	1	36	24
21	2	Cefotaxime	<=	0.25	0.03	0.12	1	36	24
21	2	Cefoxitin	=	4	2	8	1	36	24
21	2	Ceftazidime	<=	0.25	0.06	0.5	1	36	24
21	2	Ertapenem	<=	0.015	0.004	0.016	1	36	24
21	2	Imipenem	<=	0.12	0.06	0.25	1	36	24
21	2	Meropenem	<=	0.03	0.008	0.06	1	36	24

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
22	1	Ampicillin	=	2	2	8	1	36	20
22	1	Cefotaxime	<=	0.25	0.03	0.12	1	36	20
22	1	Ceftazidime	<=	0.5	0.06	0.5	1	36	20
22	1	Chloramphenicol	<=	8	2	8	1	36	20
22	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	36	20
22	1	Colistin	<=	1	0.25	2	1	36	20
22	1	Gentamicin	=	1	0.25	1	1	36	20
22	1	Meropenem	<=	0.03	0.008	0.06	1	36	20
22	1	Nalidixic acid	<=	4	1	4	1	36	20
22	1	Sulfamethoxazole	=	16	8	32	1	36	20
22	1	Tetracycline	<=	2	0.5	2	1	36	20
22	1	Tigecycline	<=	0.25	0.03	0.25	1	36	20
22	1	Trimethoprim	=	0.5	0.5	2	1	36	20
22	2	Cefepime	<=	0.06	0.016	0.12	1	36	20
22	2	Cefotaxime	<=	0.25	0.03	0.12	1	36	20
22	2	Cefoxitin	=	2	2	8	1	36	20
22	2	Ceftazidime	<=	0.25	0.06	0.5	1	36	20
22	2	Ertapenem	<=	0.015	0.004	0.016	1	36	20
22	2	Imipenem	<=	0.12	0.06	0.25	1	36	20
22	2	Meropenem	<=	0.03	0.008	0.06	1	36	20
23	1	Ampicillin	=	4	2	8	1	-	-
23	1	Cefotaxime	<=	0.25	0.03	0.12	1	-	-
23	1	Ceftazidime	<=	0.5	0.06	0.5	1	-	-
23	1	Chloramphenicol	<=	8	2	8	1	-	-
23	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	-	-
23	1	Colistin	<=	1	0.25	2	1	-	-
23	1	Gentamicin	<=	0.5	0.25	1	1	-	-
23	1	Meropenem	<=	0.03	0.008	0.06	1	-	-
23	1	Nalidixic acid	<=	4	1	4	1	-	-
23	1	Sulfamethoxazole	=	16	8	32	1	-	-
23	1	Tetracycline	<=	2	0.5	2	1	-	-
23	1	Tigecycline	<=	0.25	0.03	0.25	1	-	-
23	1	Trimethoprim	=	0.5	0.5	2	1	-	-
23	2	Cefepime	<=	0.06	0.016	0.12	1	-	-
23	2	Cefotaxime	<=	0.25	0.03	0.12	1	-	-
23	2	Cefoxitin	=	2	2	8	1	-	-
23	2	Ceftazidime	<=	0.25	0.06	0.5	1	-	-
23	2	Ertapenem	<=	0.015	0.004	0.016	1	-	-
23	2	Imipenem	<=	0.12	0.06	0.25	1	-	-
23	2	Meropenem	<=	0.03	0.008	0.06	1	-	-
25	1	Ampicillin	=	8	2	8	1	35	16-20
25	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	16-20
25	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	16-20
25	1	Chloramphenicol	<=	8	2	8	1	35	16-20
25	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	16-20
25	1	Colistin	<=	1	0.25	2	1	35	16-20
25	1	Gentamicin	=	1	0.25	1	1	35	16-20
25	1	Meropenem	<=	0.03	0.008	0.06	1	35	16-20
25	1	Nalidixic acid	<=	4	1	4	1	35	16-20
25	1	Sulfamethoxazole	<=	8	8	32	1	35	16-20
25	1	Tetracycline	<=	2	0.5	2	1	35	16-20
25	1	Tigecycline	<=	0.25	0.03	0.25	1	35	16-20
25	1	Trimethoprim	=	0.5	0.5	2	1	35	16-20
25	2	Cefepime	<=	0.06	0.016	0.12	1	35	16-20
25	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	16-20
25	2	Cefoxitin	=	2	2	8	1	35	16-20
25	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	16-20
25	2	Ertapenem	<=	0.015	0.004	0.016	1	35	16-20
25	2	Imipenem	<=	0.12	0.06	0.25	1	35	16-20
25	2	Meropenem	<=	0.03	0.008	0.06	1	35	16-20
26	1	Ampicillin	=	2	2	8	1	37	18
26	1	Cefotaxime	<=	0.25	0.03	0.12	1	37	18
26	1	Ceftazidime	<=	0.5	0.06	0.5	1	37	18
26	1	Chloramphenicol	<=	8	2	8	1	37	18
26	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37	18
26	1	Colistin	<=	1	0.25	2	1	37	18
26	1	Gentamicin	=	1	0.25	1	1	37	18
26	1	Meropenem	<=	0.03	0.008	0.06	1	37	18
26	1	Nalidixic acid	<=	4	1	4	1	37	18
26	1	Sulfamethoxazole	=	32	8	32	1	37	18
26	1	Tetracycline	<=	2	0.5	2	1	37	18
26	1	Tigecycline	<=	0.25	0.03	0.25	1	37	18
26	1	Trimethoprim	=	0.5	0.5	2	1	37	18

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
29	1	Ampicillin	=	4	2	8	1	-	-
29	1	Cefotaxime	<=	0.25	0.03	0.12	1	-	-
29	1	Ceftazidime	<=	0.5	0.06	0.5	1	-	-
29	1	Chloramphenicol	<=	8	2	8	1	-	-
29	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	-	-
29	1	Colistin	<=	1	0.25	2	1	-	-
29	1	Gentamicin	<=	0.5	0.25	1	1	-	-
29	1	Meropenem	<=	0.03	0.008	0.06	1	-	-
29	1	Nalidixic acid	<=	4	1	4	1	-	-
29	1	Sulfamethoxazole	=	32	8	32	1	-	-
29	1	Tetracycline	<=	2	0.5	2	1	-	-
29	1	Tigecycline	<=	0.25	0.03	0.25	1	-	-
29	1	Trimethoprim	=	1	0.5	2	1	-	-
29	2	Cefepime	<=	0.06	0.016	0.12	1	-	-
29	2	Cefotaxime	<=	0.25	0.03	0.12	1	-	-
29	2	Cefoxitin	=	2	2	8	1	-	-
29	2	Ceftazidime	=	0.5	0.06	0.5	1	-	-
29	2	Ertapenem	<=	0.015	0.004	0.016	1	-	-
29	2	Imipenem	<=	0.12	0.06	0.25	1	-	-
29	2	Meropenem	<=	0.03	0.008	0.06	1	-	-
30	1	Ampicillin	=	4	2	8	1	35	20
30	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	20
30	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	20
30	1	Chloramphenicol	<=	8	2	8	1	35	20
30	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	20
30	1	Colistin	<=	1	0.25	2	1	35	20
30	1	Gentamicin	<=	0.5	0.25	1	1	35	20
30	1	Meropenem	<=	0.03	0.008	0.06	1	35	20
30	1	Nalidixic acid	<=	4	1	4	1	35	20
30	1	Sulfamethoxazole	=	16	8	32	1	35	20
30	1	Tetracycline	<=	2	0.5	2	1	35	20
30	1	Tigecycline	<=	0.25	0.03	0.25	1	35	20
30	1	Trimethoprim	=	0.5	0.5	2	1	35	20
30	2	Cefepime	<=	0.06	0.016	0.12	1	35	20
30	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	20
30	2	Cefoxitin	=	2	2	8	1	35	20
30	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	20
30	2	Ertapenem	<=	0.015	0.004	0.016	1	35	20
30	2	Imipenem	<=	0.12	0.06	0.25	1	35	20
30	2	Meropenem	<=	0.03	0.008	0.06	1	35	20
32	1	Ampicillin	=	4	2	8	1	-	-
32	1	Cefotaxime	<=	0.25	0.03	0.12	1	-	-
32	1	Ceftazidime	<=	0.5	0.06	0.5	1	-	-
32	1	Chloramphenicol	<=	8	2	8	1	-	-
32	1	Ciprofloxacin	<=	0.15	0.004	0.016	1	-	-
32	1	Colistin	<=	1	0.25	2	1	-	-
32	1	Gentamicin	<=	0.5	0.25	1	1	-	-
32	1	Meropenem	<=	0.03	0.008	0.06	1	-	-
32	1	Nalidixic acid	<=	4	1	4	1	-	-
32	1	Sulfamethoxazole	=	32	8	32	1	-	-
32	1	Tetracycline	<=	2	0.5	2	1	-	-
32	1	Tigecycline	<=	0.25	0.03	0.25	1	-	-
32	1	Trimethoprim	=	0.5	0.5	2	1	-	-
32	2	Cefotaxime	<=	0.25	0.03	0.12	1	-	-
32	2	Cefoxitin	=	2	2	8	1	-	-
32	2	Ceftazidime	<=	0.25	0.06	0.5	1	-	-
32	2	Ertapenem	<=	0.015	0.004	0.016	1	-	-
32	2	Imipenem	<=	0.12	0.06	0.25	1	-	-
32	2	Meropenem	<=	0.03	0.008	0.06	1	-	-
33	1	Ampicillin	=	4	2	8	1	35	16-18
33	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	16-18
33	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	16-18
33	1	Chloramphenicol	<=	8	2	8	1	35	16-18
33	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	16-18
33	1	Colistin	<=	1	0.25	2	1	35	16-18
33	1	Gentamicin	<=	0.5	0.25	1	1	35	16-18
33	1	Meropenem	<=	0.03	0.008	0.06	1	35	16-18
33	1	Nalidixic acid	<=	4	1	4	1	35	16-18
33	1	Sulfamethoxazole	=	32	8	32	1	35	16-18
33	1	Tetracycline	<=	2	0.5	2	1	35	16-18
33	1	Tigecycline	<=	0.25	0.03	0.25	1	35	16-18
33	1	Trimethoprim	=	0.5	0.5	2	1	35	16-18
33	2	Cefepime	<=	0.06	0.016	0.12	1	35	16-18
33	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	16-18
33	2	Cefoxitin	=	2	2	8	1	35	16-18
33	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	16-18
33	2	Ertapenem	<=	0.015	0.004	0.016	1	35	16-18
33	2	Imipenem	<=	0.12	0.06	0.25	1	35	16-18
33	2	Meropenem	<=	0.03	0.008	0.06	1	35	16-18

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
34	1	Ampicillin	=	4	2	8	1	37	18-24
34	1	Cefotaxime	<=	0.25	0.03	0.12	1	37	18-24
34	1	Ceftazidime	<=	0.5	0.06	0.5	1	37	18-24
34	1	Chloramphenicol	<=	8	2	8	1	37	18-24
34	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37	18-24
34	1	Colistin	<=	1	0.25	2	1	37	18-24
34	1	Gentamicin	=	1	0.25	1	1	37	18-24
34	1	Meropenem	<=	0.03	0.008	0.06	1	37	18-24
34	1	Nalidixic acid	<=	4	1	4	1	37	18-24
34	1	Sulfamethoxazole	=	16	8	32	1	37	18-24
34	1	Tetracycline	<=	2	0.5	2	1	37	18-24
34	1	Tigecycline	<=	0.25	0.03	0.25	1	37	18-24
34	1	Trimethoprim	=	0.5	0.5	2	1	37	18-24
34	2	Cefepime	<=	0.06	0.016	0.12	1	37	18-24
34	2	Cefotaxime	<=	0.25	0.03	0.12	1	37	18-24
34	2	Cefoxitin	=	4	2	8	1	37	18-24
34	2	Ceftazidime	<=	0.25	0.06	0.5	1	37	18-24
34	2	Ertapenem	<=	0.015	0.004	0.016	1	37	18-24
34	2	Imipenem	=	0.25	0.06	0.25	1	37	18-24
34	2	Meropenem	<=	0.03	0.008	0.06	1	37	18-24
36	1	Ampicillin	=	4	2	8	1	35	18-24
36	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
36	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18-24
36	1	Chloramphenicol	<=	8	2	8	1	35	18-24
36	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18-24
36	1	Colistin	<=	1	0.25	2	1	35	18-24
36	1	Gentamicin	<=	0.5	0.25	1	1	35	18-24
36	1	Meropenem	<=	0.03	0.008	0.06	1	35	18-24
36	1	Nalidixic acid	<=	4	1	4	1	35	18-24
36	1	Sulfamethoxazole	=	16	8	32	1	35	18-24
36	1	Tetracycline	<=	2	0.5	2	1	35	18-24
36	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18-24
36	1	Trimethoprim	=	1	0.5	2	1	35	18-24
36	2	Cefepime	<=	0.06	0.016	0.12	1	35	18-24
36	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
36	2	Cefoxitin	=	4	2	8	1	35	18-24
36	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	18-24
36	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18-24
36	2	Imipenem	=	0.25	0.06	0.25	1	35	18-24
36	2	Meropenem	<=	0.03	0.008	0.06	1	35	18-24
37	1	Ampicillin	=	4	2	8	1	37	18-24
37	1	Cefotaxime	<=	0.25	0.03	0.12	1	37	18-24
37	1	Ceftazidime	<=	0.5	0.06	0.5	1	37	18-24
37	1	Chloramphenicol	<=	8	2	8	1	37	18-24
37	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37	18-24
37	1	Colistin	<=	1	0.25	2	1	37	18-24
37	1	Gentamicin	=	1	0.25	1	1	37	18-24
37	1	Meropenem	<=	0.03	0.008	0.06	1	37	18-24
37	1	Nalidixic acid	<=	4	1	4	1	37	18-24
37	1	Sulfamethoxazole	=	16	8	32	1	37	18-24
37	1	Tetracycline	<=	2	0.5	2	1	37	18-24
37	1	Tigecycline	<=	0.25	0.03	0.25	1	37	18-24
37	1	Trimethoprim	=	0.5	0.5	2	1	37	18-24
39	1	Ampicillin	=	8	2	8	1	35	18-24
39	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
39	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18-24
39	1	Chloramphenicol	<=	8	2	8	1	35	18-24
39	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18-24
39	1	Colistin	<=	1	0.25	2	1	35	18-24
39	1	Gentamicin	<=	0.5	0.25	1	1	35	18-24
39	1	Meropenem	<=	0.03	0.008	0.06	1	35	18-24
39	1	Nalidixic acid	<=	4	1	4	1	35	18-24
39	1	Sulfamethoxazole	=	32	8	32	1	35	18-24
39	1	Tetracycline	<=	2	0.5	2	1	35	18-24
39	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18-24
39	1	Trimethoprim	=	0.5	0.5	2	1	35	18-24
39	2	Cefepime	<=	0.06	0.016	0.12	1	35	18-24
39	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
39	2	Cefoxitin	=	2	2	8	1	35	18-24
39	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	18-24
39	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18-24
39	2	Imipenem	=	0.25	0.06	0.25	1	35	18-24
39	2	Meropenem	<=	0.03	0.008	0.06	1	35	18-24

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
40	1	Ampicillin	=	2	2	8	1	37	20
40	1	Cefotaxime	=	0.12	0.03	0.12	1	37	20
40	1	Ceftazidime	=	0.5	0.06	0.5	1	37	20
40	1	Chloramphenicol	=	8	2	8	1	37	20
40	1	Ciprofloxacin	=	0.015	0.004	0.016	1	37	20
40	1	Colistin	=	1	0.25	2	1	37	20
40	1	Gentamicin	=	0.5	0.25	1	1	37	20
40	1	Meropenem	=	0.03	0.008	0.06	1	37	20
40	1	Nalidixic acid	=	4	1	4	1	37	20
40	1	Sulfamethoxazole	=	16	8	32	1	37	20
40	1	Tetracycline	=	2	0.5	2	1	37	20
40	1	Tigecycline	=	0.25	0.03	0.25	1	37	20
40	1	Trimethoprim	=	0.5	0.5	2	1	37	20
40	2	Cefepime	=	0.06	0.016	0.12	1	37	20
40	2	Cefotaxime	=	0.12	0.03	0.12	1	37	20
40	2	Cefoxitin	=	2	2	8	1	37	20
40	2	Ceftazidime	=	0.5	0.06	0.5	1	37	20
40	2	Ertapenem	=	0.015	0.004	0.016	1	37	20
40	2	Imipenem	=	0.12	0.06	0.25	1	37	20
40	2	Meropenem	=	0.03	0.008	0.06	1	37	20
41	1	Ampicillin	=	2	2	8	1	37	20
41	1	Cefotaxime	<=	0.25	0.03	0.12	1	37	20
41	1	Ceftazidime	<=	0.5	0.06	0.5	1	37	20
41	1	Chloramphenicol	<=	8	2	8	1	37	20
41	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37	20
41	1	Colistin	<=	1	0.25	2	1	37	20
41	1	Gentamicin	=	1	0.25	1	1	37	20
41	1	Meropenem	<=	0.03	0.008	0.06	1	37	20
41	1	Nalidixic acid	<=	4	1	4	1	37	20
41	1	Sulfamethoxazole	=	16	8	32	1	37	20
41	1	Tetracycline	<=	2	0.5	2	1	37	20
41	1	Tigecycline	<=	0.25	0.03	0.25	1	37	20
41	1	Trimethoprim	<=	0.5	0.5	2	1	37	20
41	2	Cefepime	<=	0.06	0.016	0.12	1	37	20
41	2	Cefotaxime	<=	0.25	0.03	0.12	1	37	20
41	2	Cefoxitin	=	2	2	8	1	37	20
41	2	Ceftazidime	<=	0.5	0.06	0.5	1	37	20
41	2	Ertapenem	<=	0.015	0.004	0.016	1	37	20
41	2	Imipenem	<=	0.12	0.06	0.25	1	37	20
41	2	Meropenem	<=	0.03	0.008	0.06	1	37	20
42	1	Ampicillin	=	8	2	8	1	37	24
42	1	Cefotaxime	<=	0.25	0.03	0.12	1	37	24
42	1	Ceftazidime	<=	0.5	0.06	0.5	1	37	24
42	1	Chloramphenicol	<=	8	2	8	1	37	24
42	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37	24
42	1	Colistin	<=	1	0.25	2	1	37	24
42	1	Gentamicin	<=	0.5	0.25	1	1	37	24
42	1	Meropenem	<=	0.03	0.008	0.06	1	37	24
42	1	Nalidixic acid	<=	4	1	4	1	37	24
42	1	Sulfamethoxazole	=	32	8	32	1	37	24
42	1	Tetracycline	<=	2	0.5	2	1	37	24
42	1	Tigecycline	<=	0.25	0.03	0.25	1	37	24
42	1	Trimethoprim	=	1	0.5	2	1	37	24
42	2	Cefepime	<=	0.06	0.016	0.12	1	37	24
42	2	Cefotaxime	<=	0.25	0.03	0.12	1	37	24
42	2	Cefoxitin	=	2	2	8	1	37	24
42	2	Ceftazidime	<=	0.25	0.06	0.5	1	37	24
42	2	Ertapenem	<=	0.015	0.004	0.016	1	37	24
42	2	Imipenem	<=	0.12	0.06	0.25	1	37	24
42	2	Meropenem	<=	0.03	0.008	0.06	1	37	24
45	1	Ampicillin	=	4	2	8	1	36/37	20
45	1	Cefotaxime	<=	0.25	0.03	0.12	1	36/37	20
45	1	Ceftazidime	<=	0.5	0.06	0.5	1	36/37	20
45	1	Chloramphenicol	<=	8	2	8	1	36/37	20
45	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	36/37	20
45	1	Colistin	<=	1	0.25	2	1	36/37	20
45	1	Gentamicin	<=	0.5	0.25	1	1	36/37	20
45	1	Meropenem	<=	0.03	0.008	0.06	1	36/37	20
45	1	Nalidixic acid	<=	4	1	4	1	36/37	20
45	1	Sulfamethoxazole	<=	8	8	32	1	36/37	20
45	1	Tetracycline	<=	2	0.5	2	1	36/37	20
45	1	Tigecycline	<=	0.25	0.03	0.25	1	36/37	20
45	1	Trimethoprim	=	1	0.5	2	1	36/37	20
45	2	Cefepime	<=	0.06	0.016	0.12	1	36/37	20
45	2	Cefotaxime	<=	0.25	0.03	0.12	1	36/37	20
45	2	Cefoxitin	=	4	2	8	1	36/37	20
45	2	Ceftazidime	<=	0.25	0.06	0.5	1	36/37	20
45	2	Ertapenem	<=	0.015	0.004	0.016	1	36/37	20
45	2	Imipenem	<=	0.12	0.06	0.25	1	36/37	20
45	2	Meropenem	<=	0.03	0.008	0.06	1	36/37	20

Lab no.	Panel	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Temperature	Time
56	1	Ampicillin	=	2	2	8	1	35	20
56	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	20
56	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	20
56	1	Chloramphenicol	<=	8	2	8	1	35	20
56	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	20
56	1	Colistin	<=	1	0.25	2	1	35	20
56	1	Gentamicin	<=	0.5	0.25	1	1	35	20
56	1	Meropenem	<=	0.03	0.008	0.06	1	35	20
56	1	Nalidixic acid	<=	4	1	4	1	35	20
56	1	Sulfamethoxazole	=	32	8	32	1	35	20
56	1	Tetracycline	<=	2	0.5	2	1	35	20
56	1	Tigecycline	<=	0.25	0.03	0.25	1	35	20
56	1	Trimethoprim	=	0.5	0.5	2	1	35	20
56	2	Cefepime	<=	0.06	0.016	0.12	1	35	20
56	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	20
56	2	Cefoxitin	=	4	2	8	1	35	20
56	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	20
56	2	Ertapenem	<=	0.015	0.004	0.016	1	35	20
56	2	Imipenem	<=	0.12	0.06	0.25	1	35	20
56	2	Meropenem	<=	0.03	0.008	0.06	1	35	20
59	1	Ampicillin	=	2	2	8	1	35	18-24
59	1	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
59	1	Ceftazidime	<=	0.5	0.06	0.5	1	35	18-24
59	1	Chloramphenicol	<=	8	2	8	1	35	18-24
59	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	35	18-24
59	1	Colistin	<=	1	0.25	2	1	35	18-24
59	1	Gentamicin	<=	0.5	0.25	1	1	35	18-24
59	1	Meropenem	<=	0.03	0.008	0.06	1	35	18-24
59	1	Nalidixic acid	<=	4	1	4	1	35	18-24
59	1	Tetracycline	<=	2	0.5	2	1	35	18-24
59	1	Tigecycline	<=	0.25	0.03	0.25	1	35	18-24
59	1	Trimethoprim	<=	**0.25**	0.5	2	**0**	35	18-24
59	2	Cefepime	<=	0.06	0.016	0.12	1	35	18-24
59	2	Cefotaxime	<=	0.25	0.03	0.12	1	35	18-24
59	2	Cefoxitin	=	2	2	8	1	35	18-24
59	2	Ceftazidime	<=	0.25	0.06	0.5	1	35	18-24
59	2	Ertapenem	<=	0.015	0.004	0.016	1	35	18-24
59	2	Imipenem	<=	0.12	0.06	0.25	1	35	18-24
59	2	Meropenem	<=	0.03	0.008	0.06	1	35	18-24
60	1	Ampicillin	=	4	2	8	1	36±1	18-20
60	1	Cefotaxime	=	**0.25**	0.03	0.12	**0**	36±1	18-20
60	1	Ceftazidime	=	0.5	0.06	0.5	1	36±1	18-20
60	1	Chloramphenicol	=	8	2	8	1	36±1	18-20
60	1	Ciprofloxacin	=	0.015	0.004	0.016	1	36±1	18-20
60	1	Colistin	=	1	0.25	2	1	36±1	18-20
60	1	Gentamicin	=	0.5	0.25	1	1	36±1	18-20
60	1	Meropenem	=	0.03	0.008	0.06	1	36±1	18-20
60	1	Nalidixic acid	=	4	1	4	1	36±1	18-20
60	1	Sulfamethoxazole	=	8	8	32	1	36±1	18-20
60	1	Tetracycline	=	2	0.5	2	1	36±1	18-20
60	1	Tigecycline	=	0.25	0.03	0.25	1	36±1	18-20
60	1	Trimethoprim	=	0.5	0.5	2	1	36±1	18-20
60	2	Cefepime	<=	0.06	0.016	0.12	1	36±1	18-20
60	2	Cefotaxime	<=	0.25	0.03	0.12	1	36±1	18-20
60	2	Cefoxitin	=	4	2	8	1	36±1	18-20
60	2	Ceftazidime	<=	0.25	0.06	0.5	1	36±1	18-20
60	2	Ertapenem	<=	0.015	0.004	0.016	1	36±1	18-20
60	2	Imipenem	=	0.25	0.06	0.25	1	36±1	18-20
60	2	Meropenem	<=	0.03	0.008	0.06	1	36±1	18-20
64	1	Ampicillin	=	2	2	8	1	37	18
64	1	Cefotaxime	<=	0.25	0.03	0.12	1	37	18
64	1	Ceftazidime	<=	0.5	0.06	0.5	1	37	18
64	1	Chloramphenicol	<=	8	2	8	1	37	18
64	1	Ciprofloxacin	<=	0.015	0.004	0.016	1	37	18
64	1	Colistin	<=	1	0.25	2	1	37	18
64	1	Gentamicin	=	1	0.25	1	1	37	18
64	1	Meropenem	<=	0.03	0.008	0.06	1	37	18
64	1	Nalidixic acid	<=	4	1	4	1	37	18
64	1	Sulfamethoxazole	=	32	8	32	1	37	18
64	1	Tetracycline	<=	2	0.5	2	1	37	18
64	1	Tigecycline	<=	0.25	0.03	0.25	1	37	18
64	1	Trimethoprim	=	1	0.5	2	1	37	18

Test results from the reference strain *C. jejuni* ATCC 33560

Lab no.	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Method	36-37ºC/48h	42ºC/24h
2	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
2	Erythromycin ERY	=	1	0.5	2	1	MIC	X	
2	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
2	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
2	Tetracycline TET	=	2	0.25	2	1	MIC	X	
4	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
4	Erythromycin ERY	=	2	0.5	2	1	MIC	X	
4	Gentamicin GEN	=	0.5	0.5	2	1	MIC	X	
4	Nalidixic acid NAL	=	16	4	16	1	MIC	X	
4	Tetracycline TET	=	4	0.25	2	**0**	MIC	X	
6	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
6	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
6	Gentamicin GEN	=	1	0.25	2	1	MIC		X
6	Nalidixic acid NAL	=	8	4	16	1	MIC		X
6	Tetracycline TET	=	1	0.25	1	1	MIC		X
9	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
9	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
9	Gentamicin GEN	=	0.5	0.5	2	1	MIC	X	
9	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
9	Tetracycline TET	=	1	0.25	2	1	MIC	X	
11	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
11	Erythromycin ERY	=	2	0.25	2	1	MIC		X
11	Gentamicin GEN	=	1	0.25	2	1	MIC		X
11	Nalidixic acid NAL	=	8	4	16	1	MIC		X
11	Tetracycline TET	=	1	0.25	1	1	MIC		X
12	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
12	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
12	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
12	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
12	Tetracycline TET	<=	0.5	0.25	2	1	MIC	X	
14	Ciprofloxacin CIP	<=	0.125	0.03	0.125	1	MIC		X
14	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
14	Gentamicin GEN	=	0.5	0.25	2	1	MIC		X
14	Nalidixic acid NAL	=	4	4	16	1	MIC		X
14	Tetracycline TET	<=	0.5	0.25	1	1	MIC		X
17	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
17	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
17	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
17	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
17	Tetracycline TET	<=	0.5	0.25	2	1	MIC	X	
18	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
18	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
18	Gentamicin GEN	=	0.5	0.25	2	1	MIC		X
18	Nalidixic acid NAL	=	4	4	16	1	MIC		X
18	Tetracycline TET	<=	0.5	0.25	1	1	MIC		X
19	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
19	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
19	Gentamicin GEN	=	0.5	0.25	2	1	MIC		X
19	Nalidixic acid NAL	=	8	4	16	1	MIC		X
19	Tetracycline TET	<=	0.5	0.25	1	1	MIC		X
20	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
20	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
20	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
20	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
20	Tetracycline TET	=	1	0.25	2	1	MIC	X	
21	Ciprofloxacin CIP	=	0.12	0.03	0.125	1	MIC		X
21	Erythromycin ERY	=	1	0.25	2	1	MIC		X
21	Gentamicin GEN	=	0.5	0.25	2	1	MIC		X
21	Nalidixic acid NAL	=	4	4	16	1	MIC		X
21	Tetracycline TET	=	0.5	0.25	1	1	MIC		X
22	Ciprofloxacin CIP	<=	0.125	0.03	0.125	1	MIC		X
22	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
22	Gentamicin GEN	=	0.5	0.25	2	1	MIC		X
22	Nalidixic acid NAL	=	4	4	16	1	MIC		X
22	Tetracycline TET	=	1	0.25	1	1	MIC		X
23	Ciprofloxacin CIP	=	0.06	0.03	0.125	1	MIC		X
23	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
23	Gentamicin GEN	=	1	0.25	2	1	MIC		X
23	Nalidixic acid NAL	=	4	4	16	1	MIC		X
23	Tetracycline TET	=	1	0.25	1	1	MIC		X
25	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
25	Erythromycin ERY	=	2	0.5	2	1	MIC	X	
25	Gentamicin GEN	=	0.25	0.5	2	**0**	MIC	X	
25	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
25	Tetracycline TET	=	2	0.25	2	1	MIC	X	

Lab no.	Antimicrobial	Operator	Value	Low limit	High limit	Mark	Method	36-37ºC/48h	42ºC/24h
26	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
26	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
26	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
26	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
26	Tetracycline TET	=	1	0.25	2	1	MIC	X	
30	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
30	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
30	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
30	Nalidixic acid NAL	=	4	4	16	1	MIC	X	
30	Tetracycline TET	=	2	0.25	2	1	MIC	X	
32	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
32	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
32	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
32	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
32	Tetracycline TET	<=	0.5	0.25	2	1	MIC	X	
33	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
33	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
33	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
33	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
33	Tetracycline TET	<=	0.5	0.25	2	1	MIC	X	
34	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
34	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
34	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
34	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
34	Tetracycline TET	=	1	0.25	2	1	MIC	X	
36	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
36	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
36	Gentamicin GEN	=	0.5	0.25	2	1	MIC		X
36	Nalidixic acid NAL	=	8	4	16	1	MIC		X
36	Tetracycline TET	=	1	0.25	1	1	MIC		X
37	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
37	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
37	Gentamicin GEN	=	0.25	0.5	2	**0**	MIC	X	
37	Nalidixic acid NAL	=	4	4	16	1	MIC	X	
37	Tetracycline TET	<=	0.5	0.25	2	1	MIC	X	
39	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
39	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
39	Gentamicin GEN	=	0.5	0.25	2	1	MIC		X
39	Nalidixic acid NAL	=	4	4	16	1	MIC		X
39	Tetracycline TET	<=	0.5	0.25	1	1	MIC		X
40	Ciprofloxacin CIP	=	0.12	0.03	0.125	1	MIC		X
40	Erythromycin ERY	=	1	0.25	2	1	MIC		X
40	Nalidixic acid NAL	=	8	4	16	1	MIC		X
40	Tetracycline TET	=	0.5	0.25	1	1	MIC		X
41	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
41	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
41	Gentamicin GEN	=	1	0.25	2	1	MIC		X
41	Nalidixic acid NAL	=	8	4	16	1	MIC		X
41	Tetracycline TET	=	1	0.25	1	1	MIC		X
42	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
42	Erythromycin ERY	=	2	0.5	2	1	MIC	X	
42	Gentamicin GEN	=	0.25	0.5	2	**0**	MIC	X	
42	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
42	Tetracycline TET	=	2	0.25	2	1	MIC	X	
45	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
45	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
45	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
45	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
45	Tetracycline TET	=	2	0.25	2	1	MIC	X	
56	Ciprofloxacin CIP	<=	0.12	0.03	0.125	1	MIC		X
56	Erythromycin ERY	<=	1	0.25	2	1	MIC		X
56	Gentamicin GEN	=	0.25	0.25	2	1	MIC		X
56	Nalidixic acid NAL	=	4	4	16	1	MIC		X
56	Tetracycline TET	<=	0.5	0.25	1	1	MIC		X
59	Ciprofloxacin CIP	<=	0.12	0.06	0.25	1	MIC	X	
59	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
59	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
59	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
59	Tetracycline TET	=	1	0.25	2	1	MIC	X	
60	Ciprofloxacin CIP	=	0.25	0.06	0.25	1	MIC	X	
60	Erythromycin ERY	<=	1	0.5	2	1	MIC	X	
60	Gentamicin GEN	=	1	0.5	2	1	MIC	X	
60	Nalidixic acid NAL	=	8	4	16	1	MIC	X	
60	Tetracycline TET	=	1	0.25	2	1	MIC	X	

MIC: Microbroth dilution

Salmonella - expected and obtained interpretation

Antimicrobial	Strain	Panel	Expected	% R	% S	No. correct	No. incorrect
Ampicillin AMP	EURL S-13.1	Panel 1	R	100	0	32	0
	EURL S-13.2	Panel 1	R	100	0	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	R	96.9	3.1	31	1
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	R	100	0	32	0
	EURL S-13.7	Panel 1	R	100	0	32	0
	EURL S-13.8	Panel 1	R	100	0	32	0
Azithromycin AZI	EURL S-13.1	Panel 1	S	0	100	24	0
	EURL S-13.2	Panel 1	R	100	0	24	0
	EURL S-13.3	Panel 1	S	0	100	24	0
	EURL S-13.4	Panel 1	S	0	100	24	0
	EURL S-13.5	Panel 1	S	0	100	24	0
	EURL S-13.6	Panel 1	R	95.8	4.2	23	1
	EURL S-13.7	Panel 1	S	0	100	24	0
	EURL S-13.8	Panel 1	R	100	0	24	0
Cefotaxime FOT	EURL S-13.1	Panel 1	R	100	0	32	0
	EURL S-13.2	Panel 1	S	0	100	31	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	R	100	0	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	S	12.5	87.5	28	4
	EURL S-13.7	Panel 1	R	100	0	32	0
	EURL S-13.8	Panel 1	R	100	0	32	0
	EURL S-13.1	Panel 2	R	100	0	32	0
	EURL S-13.4	Panel 2	R	100	0	32	0
	EURL S-13.6	Panel 2	S	9.7	90.3	28	3
	EURL S-13.7	Panel 2	R	100	0	32	0
	EURL S-13.8	Panel 2	R	100	0	32	0
Cefoxitin FOX	EURL S-13.1	Panel 2	S	3.1	96.9	31	1
	EURL S-13.4	Panel 2	S	3.1	96.9	31	1
	EURL S-13.6	Panel 2	S	12.9	87.1	27	4
	EURL S-13.7*	Panel 2*	S*	35.5*	64.5*	20*	11*
	EURL S-13.8	Panel 2	S	3.1	96.9	31	1
Ceftazidime TAZ	EURL S-13.1*	Panel 1*	R*	61.3*	38.7*	19*	12*
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	S	0	100	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	S	0	100	32	0
	EURL S-13.7	Panel 1	R	100	0	32	0
	EURL S-13.8	Panel 1	R	100	0	32	0
	EURL S-13.1*	Panel 2*	R*	62.5*	37.5*	20*	12*
	EURL S-13.4	Panel 2	S	3.1	96.9	31	1
	EURL S-13.6	Panel 2	S	0	100	31	0
	EURL S-13.7	Panel 2	R	100	0	32	0
	EURL S-13.8	Panel 2	R	100	0	32	0
Chloramphenicol CHL	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	3.1	96.9	31	1
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	S	0	100	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	S	0	100	32	0
	EURL S-13.7	Panel 1	R	100	0	32	0
	EURL S-13.8	Panel 1	S	0	100	32	0

Antimicrobial	Strain	Panel	Expected	% R	% S	No. correct	No. incorrect
Ciprofloxacin CIP	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	R	93.8	6.2	30	2
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	R	96.9	3.1	31	1
	EURL S-13.7	Panel 1	S	0	100	32	0
	EURL S-13.8	Panel 1	S	0	100	32	0
Colistin COL	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	S	0	100	32	0
	EURL S-13.5	Panel 1	R	90.3	9.7	28	3
	EURL S-13.6	Panel 1	S	0	100	32	0
	EURL S-13.7	Panel 1	S	0	100	32	0
	EURL S-13.8	Panel 1	S	0	100	32	0
Ertapenem ETP	EURL S-13.1	Panel 2	S	0	100	31	0
	EURL S-13.4	Panel 2	S	0	100	32	0
	EURL S-13.6	Panel 2	R	100	0	31	0
	EURL S-13.7	Panel 2	S	3.1	96.9	31	1
	EURL S-13.8	Panel 2	S	0	100	32	0
Gentamicin GEN	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	S	0	100	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	S	0	100	32	0
	EURL S-13.7	Panel 1	R	100	0	32	0
	EURL S-13.8	Panel 1	R	100	0	32	0
Imipenem IMI	EURL S-13.1	Panel 2	S	0	100	32	0
	EURL S-13.4	Panel 2	S	0	100	32	0
	EURL S-13.6	Panel 2	S	16.1	83.9	26	5
	EURL S-13.7	Panel 2	S	0	100	32	0
	EURL S-13.8	Panel 2	S	0	100	32	0
Meropenem MER	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	S	0	100	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	R	90.6	9.4	29	3
	EURL S-13.7	Panel 1	S	0	100	32	0
	EURL S-13.8	Panel 1	S	0	100	32	0
	EURL S-13.1	Panel 2	S	0	100	32	0
	EURL S-13.4	Panel 2	S	0	100	32	0
	EURL S-13.6	Panel 2	R	93.5	6.5	29	2
	EURL S-13.7	Panel 2	S	0	100	32	0
	EURL S-13.8	Panel 2	S	0	100	32	0
Nalidixic acid NAL	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	R	100	0	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	R	100	0	32	0
	EURL S-13.7	Panel 1	S	0	100	32	0
	EURL S-13.8	Panel 1	S	0	100	32	0

Antimicrobial	Strain	Panel	Expected	% R	% S	No. correct	No. incorrect
Sulfamethoxazole SMX	EURL S-13.1	Panel 1	R	100	0	31	0
	EURL S-13.2	Panel 1	S	0	100	31	0
	EURL S-13.3	Panel 1	S	12.9	87.1	27	4
	EURL S-13.4	Panel 1	S	0	100	31	0
	EURL S-13.5	Panel 1	R	100	0	31	0
	EURL S-13.6	Panel 1	S	3.2	96.8	30	1
	EURL S-13.7	Panel 1	R	100	0	31	0
	EURL S-13.8	Panel 1	R	96.8	3.2	30	1
Temocillin TRM	EURL S-13.1	Panel 2	S	0	100	24	0
	EURL S-13.4	Panel 2	S	0	100	25	0
	EURL S-13.6	Panel 2	R	100	0	25	0
	EURL S-13.7	Panel 2	R	95.8	4.2	23	1
	EURL S-13.8	Panel 2	S	0	100	24	0
Tetracycline TET	EURL S-13.1	Panel 1	R	100	0	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	R	100	0	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	S	0	100	32	0
	EURL S-13.7	Panel 1	R	100	0	32	0
	EURL S-13.8	Panel 1	S	0	100	32	0
Tigecycline TGC	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	S	0	100	32	0
	EURL S-13.5	Panel 1	S	0	100	32	0
	EURL S-13.6	Panel 1	S	0	100	32	0
	EURL S-13.7	Panel 1	S	0	100	32	0
	EURL S-13.8	Panel 1	S	0	100	32	0
Trimethoprim TMP	EURL S-13.1	Panel 1	S	0	100	32	0
	EURL S-13.2	Panel 1	S	0	100	32	0
	EURL S-13.3	Panel 1	S	0	100	32	0
	EURL S-13.4	Panel 1	S	0	100	31	0
	EURL S-13.5	Panel 1	R	96.9	3.1	31	1
	EURL S-13.6	Panel 1	S	0	100	32	0
	EURL S-13.7	Panel 1	R	100	0	32	0
	EURL S-13.8	Panel 1	R	96.9	3.1	31	1

**Strain/antimicrobial-combination excluded from the evaluation*

Campylobacter - expected and obtained interpretation

Antimicrobial	Strain	Expected	% R	% S	No. correct	No. incorrect
Ciprofloxacin, CIP	EURL C-13.1	R	100	0	31	0
	EURL C-13.2	S	0	100	31	0
	EURL C-13.3	R	100	0	30	0
	EURL C-13.4	R	100	0	31	0
	EURL C-13.5	S	0	100	31	0
	EURL C-13.6	R	97	3	29	1
	EURL C-13.7	S	0	100	31	0
	EURL C-13.8	R	100	0	31	0
Erythromycin, ERY	EURL C-13.1	S	0	100	31	0
	EURL C-13.2	R	100	0	31	0
	EURL C-13.3	S	7	93	28	2
	EURL C-13.4	R	100	0	31	0
	EURL C-13.5	S	0	100	31	0
	EURL C-13.6	S	0	100	31	0
	EURL C-13.7	R	100	0	31	0
	EURL C-13.8	S	0	100	31	0
Gentamicin, GEN	EURL C-13.1	S	0	100	31	0
	EURL C-13.2	S	0	100	31	0
	EURL C-13.3	S	0	100	30	0
	EURL C-13.4	R	100	0	31	0
	EURL C-13.5	S	0	100	31	0
	EURL C-13.6	S	0	100	31	0
	EURL C-13.7	S	0	100	31	0
	EURL C-13.8	S	0	100	31	0
Nalidixic acid, NAL	EURL C-13.1	R	97	3	30	1
	EURL C-13.2	S	0	100	31	0
	EURL C-13.3	R	100	0	30	0
	EURL C-13.4	R	100	0	31	0
	EURL C-13.5	S	0	100	31	0
	EURL C-13.6	S	3	97	29	1
	EURL C-13.7	S	0	100	31	0
	EURL C-13.8	R	100	0	31	0
Streptomycin, STR	EURL C-13.1	S	0	100	31	0
	EURL C-13.2	S	0	100	31	0
	EURL C-13.3	R	100	0	30	0
	EURL C-13.4	R	100	0	31	0
	EURL C-13.5	S	0	100	31	0
	EURL C-13.6	S	0	100	31	0
	EURL C-13.7	R	100	0	31	0
	EURL C-13.8	R	100	0	31	0
Tetracycline, TET	EURL C-13.1	R	97	3	30	1
	EURL C-13.2	S	3	97	30	1
	EURL C-13.3	R	100	0	30	0
	EURL C-13.4	R	100	0	31	0
	EURL C-13.5	S	0	100	31	0
	EURL C-13.6	S	3	97	30	1
	EURL C-13.7	S	0	100	31	0
	EURL C-13.8	S	0	100	31	0

Deviations - *Salmonella*

Lab no.	Strain	Panel	Antimicrobial	Obtained MIC value	Expected MIC-value	Obtained interpretation	Expected interpretation
4	EURL S-13.3	1	Sulfamethoxazole SMX	512	32	R	S
4	EURL S-13.4	1	Ciprofloxacin CIP	= 0.25	0.5	S	R
4	EURL S-13.6	1	Ciprofloxacin CIP	8	8	S	R
4	EURL S-13.6	1	Meropenem MER	= 0.12	0.5	S	R
4	EURL S-13.6	2	Meropenem MER	= 0.12	0.5	S	R
4	EURL S-13.6					Other phenotype	Presumptive carbapenemase
4	EURL S-13.7	2	Temocillin TRM	32	64	S	R
4	EURL S-13.8	1	Sulfamethoxazole SMX	> 1024	> 1024	S	R
6	EURL S-13.6	1	Cefotaxime FOT	1	0.5	R	S
6	EURL S-13.6	2	Cefotaxime FOT	1	0.5	R	S
6	EURL S-13.6	2	Cefoxitin FOX	16	8	R	S
6	EURL S-13.6	2	Imipenem IMI	2	1	R	S
12	EURL S-13.4	2	Ceftazidime TAZ	4	1	R	S
17	EURL S-13.6	2	Imipenem IMI	2	1	R	S
18	EURL S-13.3	1	Sulfamethoxazole SMX	> 1024	32	R	S
19	EURL S-13.1	2	Cefoxitin FOX	8	8	R	S
19	EURL S-13.4	2	Cefoxitin FOX	4	4	R	S
19	EURL S-13.6	2	Cefoxitin FOX	8	8	R	S
19	EURL S-13.7	2	Ertapenem ETP	= 0.12	0.06	R	S
19	EURL S-13.8	2	Cefoxitin FOX	4	4	R	S
22	EURL S-13.6	1	Cefotaxime FOT	2	0.5	R	S
22	EURL S-13.6	2	Cefotaxime FOT	2	0.5	R	S
22	EURL S-13.6	2	Cefoxitin FOX	16	8	R	S
25	EURL S-13.6	2	Imipenem IMI	2	1	R	S
26	EURL S-13.5	1	Colistin COL	<= 1	4	S	R
26	EURL S-13.6	1	Azithromycin AZI	16	32	S	R
26	EURL S-13.6	2	Meropenem MER	= 0.12	0.5	S	R
26	EURL S-13.6					Other phenotype	Presumptive carbapenemase
32	EURL S-13.3	1	Sulfamethoxazole SMX	> 1024	32	R	S
36	EURL S-13.6	2	Cefotaxime FOT	1	0.5	R	S
37	EURL S-13.6	2	Imipenem IMI	2	1	R	S
39	EURL S-13.5	1	Colistin COL	2	4	S	R
39	EURL S-13.5	1	Trimethoprim TMP	> 32	> 32	S	R
39	EURL S-13.5					Presumptive ESBL	No ESBL, AmpC- or carbapenemase
39	EURL S-13.6	1	Sulfamethoxazole SMX	1024	<= 8	R	S
40	EURL S-13.4	1	Ampicillin AMP	> 64	> 64	S	R
40	EURL S-13.8	1	Trimethoprim TMP	<= 0.25	> 32	S	R
41	EURL S-13.6	2	Cefoxitin FOX	16	8	R	S
41	EURL S-13.6	2	Imipenem IMI	2	1	R	S
42	EURL S-13.6	1	Cefotaxime FOT	1	0.5	R	S
42	EURL S-13.6	1	Meropenem MER	= 0.12	0.5	S	R
45	EURL S-13.5	1	Colistin COL	2	4	S	R
59	EURL S-13.3	1	Sulfamethoxazole SMX	> 1024	32	R	S
59	EURL S-13.4	1	Ciprofloxacin CIP	= 0.25	0.5	S	R
60	EURL S-13.6	1	Cefotaxime FOT	= 0.5	0.5	R	S
64	EURL S-13.2	1	Chloramphenicol CHL	32	<= 8	R	S
64	EURL S-13.4					No ESBL, AmpC- or carbapenemase	Presumptive ESBL
64	EURL S-13.5					Other phenotype	No ESBL, AmpC- or carbapenemase
64	EURL S-13.6	1	Meropenem MER	= 0.12	0.5	S	R
64	EURL S-13.6					No ESBL, AmpC- or carbapenemase	Presumptive carbapenemase

Deviations - *Campylobacter*

Lab no.	Strain	Antimicrobial	Obtained MIC value	Expected MIC-value	Obtained interpretation	Expected interpretation
14	EURL C-13.1	Nalidixic acid NAL	2	64	S	R
14	EURL C-13.1	Tetracycline TET	<= 0.5	32	S	R
14	EURL C-13.6	Nalidixic acid NAL	64	<= 1	R	S
14	EURL C-13.6	Tetracycline TET	64	<= 0.5	R	S
18	EURL C-13.3	Erythromycin ERY	16	4	R	S
30	EURL C-13.2	Tetracycline TET	4	2	R	S
32	EURL C-13.6	Ciprofloxacin CIP	<= 0.12	8	S	R
42	EURL C-13.3	Erythromycin ERY	16	4	R	S

Genotypic characterization (optional); genes detected in the ESBL-, AmpC, and carbapenemase producing *Salmonella* strains

Labno	Strain	Genetype	Gene number	Method	Reference	Primer 5 3	Primer 3 5
1	EURL S-13.1	CTX	M-8	Whole genome sequenced			
1	EURL S-13.2	TEM	-1B	Whole genome sequenced			
1	EURL S-13.4	CTX	M-9	Whole genome sequenced			
1	EURL S-13.4	TEM	-1B	Whole genome sequenced			
1	EURL S-13.6	OXA	-48	Whole genome sequenced			
1	EURL S-13.7	CTX	M-15	Whole genome sequenced			
1	EURL S-13.7	SHV	-12	Whole genome sequenced			
1	EURL S-13.8	CTX	M-3	Whole genome sequenced			
1	EURL S-13.8	TEM	-1B	Whole genome sequenced			
2	EURL S-13.1	CTX	M-8	Whole genome sequenced	ResFinder 3.1		
2	EURL S-13.4	CTX	M-9	Whole genome sequenced	ResFinder 3.1		
2	EURL S-13.4	TEM	-1B	Whole genome sequenced	ResFinder 3.1		
2	EURL S-13.6	OXA	-48	Whole genome sequenced	ResFinder 3.1		
2	EURL S-13.7	CTX	M-15	Whole genome sequenced	ResFinder 3.1		
2	EURL S-13.7	SHV	-12	Whole genome sequenced	ResFinder 3.1		
2	EURL S-13.8	CTX	M-3	Whole genome sequenced	ResFinder 3.1		
2	EURL S-13.8	TEM	-1B	Whole genome sequenced	ResFinder 3.1		
9	EURL S-13.1	CTX	M-8	PCR (published)	JAC 2010;65;490-495		
9	EURL S-13.4	CTX	M-9	PCR (published)	JAC 2010;65;490-495		
9	EURL S-13.4	TEM	-1	PCR (published)	JAC 2010;65;490-495		
9	EURL S-13.6	OXA	-48	PCR (published)	JAC 2010;65;490-495		
9	EURL S-13.7	CTX	M-15	PCR (published)	JAC 2010;65;490-495		
9	EURL S-13.7	SHV	-12	PCR (published)	JAC 2010;65;490-495		
9	EURL S-13.8	CTX	M-3	PCR (published)	JAC 2010;65;490-495		
9	EURL S-13.8	TEM	-1	PCR (published)	JAC 2010;65;490-495		
17	EURL S-13.1	CTX	M-8	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.2	TEM	-1B	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.4	CTX	M-9	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.4	TEM	-1B	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.6	OXA	-48	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.7	CTX	M-15	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.7	SHV	-12	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.8	CTX	M-3	Whole genome sequenced	ResFinder 3.1		
17	EURL S-13.8	TEM	-1B	Whole genome sequenced	ResFinder 3.1		
20	EURL S-13.1	CTX	M-8	Whole genome sequenced	CTX-M-8 AF189721		
20	EURL S-13.2	TEM	-1B	Whole genome sequenced	bla TEM-1B AY458016		
20	EURL S-13.4	CTX	M-9	Whole genome sequenced	CTX-M-9 AF174129		
20	EURL S-13.4	TEM	-1B	Whole genome sequenced	blaTEM-1B AY458016		
20	EURL S-13.6	OXA	-48	Whole genome sequenced	bla OXA-48 AY236073		
20	EURL S-13.7	CTX	M-15	Whole genome sequenced	bla CTX-M-15 AY044436		
20	EURL S-13.7	SHV	-12	Whole genome sequenced	blaSHV-12 KF976405		
20	EURL S-13.8	CTX	M-3	Whole genome sequenced	bla CTX-M-3 Y10278		
20	EURL S-13.8	TEM	-1B	Whole genome sequenced	blaTEM 1B AY458016		
21	EURL S-13.4	CTX		PCR (published)			

Labno	Strain	Genetype	Gene number	Method	Reference	Primer 5 3	Primer 3 5
21	EURL S-13.6	OXA	-48	PCR (published)			
21	EURL S-13.7	CTX		PCR (published)			
21	EURL S-13.7	SHV		PCR (published)			
21	EURL S-13.8	CTX		PCR (published)			
22	EURL S-13.1	CTX		Whole genome sequenced			
22	EURL S-13.4	CTX		Whole genome sequenced			
22	EURL S-13.4	TEM		Whole genome sequenced			
22	EURL S-13.4	TEM		PCR (published)	Kim et al., 2009	AGT GCT GCC ATA ACC ATG AGT G	CTG ACT CCC CGT CGT GTA GAT A
22	EURL S-13.5	CTX		PCR (published)	Kim et al., 2009	GAC AAA GAG AGT GCA ACG GAT G	TCA GTG CGA TCC AGA CGA AA
22	EURL S-13.6	OXA		Whole genome sequenced			
22	EURL S-13.7	CTX		Whole genome sequenced			
22	EURL S-13.7	CTX		PCR (published)	Kim et al., 2009	TCC AGA ATA AGG AAT CCC ATG G	TGC TTT ACC CAG CGT CAG AT
22	EURL S-13.7	SHV		PCR (published)	Kim et al., 2009	GAT GAA CGC TTT CCC ATG ATG	CGC TGT TAT CGC TCA TGG TAA
22	EURL S-13.8	CTX		Whole genome sequenced			
22	EURL S-13.8	CTX		PCR (published)	Kim et al., 2009	TCC AGA ATA AGG AAT CCC ATG G	TGC TTT ACC CAG CGT CAG AT
22	EURL S-13.8	TEM		PCR (published)	Kim et al., 2009	AGT GCT GCC ATA ACC ATG AGT G	CTG ACT CCC CGT CGT GTA GAT A
25	EURL S-13.1	CTX	M-8	PCR (in-house)			
25	EURL S-13.4	CTX	M-9	PCR (in-house)			
25	EURL S-13.6	OXA	-48	PCR (in-house)			
25	EURL S-13.7	CTX	M-15	PCR (in-house)			
25	EURL S-13.7	SHV	-12	PCR (in-house)			
25	EURL S-13.8	CTX	M-3	PCR (in-house)			
25	EURL S-13.8	TEM	-1B	PCR (in-house)			
32	EURL S-13.1	CTX	M-8	PCR (published)	PediatrInfectDisJ28:814-818		
32	EURL S-13.1	DHA		PCR (published)			
32	EURL S-13.4	CTX	M-9	PCR (published)	PediatrInfectDisJ28:814-818		
32	EURL S-13.4	TEM	-1	PCR (published)	AntimicrAgentsChemotherap2009		
32	EURL S-13.6	OXA	-48	PCR (published)	J. Antimic.Chemothe(2009)		
32	EURL S-13.7	SHV	-12	PCR (published)	FEMSMicrobiolLett1997152:1637		
32	EURL S-13.8	TEM	-1	PCR (published)	AntimicrAgentsChemotherap2009		
33	EURL S-13.1	CTX	M-8	Whole genome sequenced	M. Hunt et al. Ariba		
33	EURL S-13.4	CTX	M-9	Whole genome sequenced	M. Hunt et al. Ariba		
33	EURL S-13.4	TEM	-1B	Whole genome sequenced	M. Hunt et al. Ariba		
33	EURL S-13.6	OXA	-48	Whole genome sequenced	M. Hunt et al. Ariba		
33	EURL S-13.7	CTX	M-15	Whole genome sequenced	M. Hunt et al. Ariba		
33	EURL S-13.7	SHV	-12	Whole genome sequenced	M. Hunt et al. Ariba		
33	EURL S-13.8	CTX	M-3	Whole genome sequenced	M. Hunt et al. Ariba		
33	EURL S-13.8	TEM	-1B	Whole genome sequenced	M. Hunt et al. Ariba		
36	EURL S-13.1	CTX		PCR (published)	Hasman et al. beta-Lactamases among extendedspectrum beta-lactamase (ESBL)-resistant Salmonella from poultry, poultry products and human patients in The Netherlands. JAC 2005 Jul;56(1):115-21	ATGTGCAGYACCAGTAARGTKATGGC	TGGGTRAARTARGTSACCAGAAYCAGCGG

Labno	Strain	Genetype	Gene number	Method	Reference	Primer 5 3	Primer 3 5
36	EURL S-13.4	CTX		PCR (published)	Hasman et al beta-Lactamases among extendedspectrum beta-lactamase (ESBL)-resistant Salmonella from poultry, poultry products and human patients in The Netherlands. JAC 2005 Jul;56(1):115-21	ATGTGCAGYACCAGTAARGTKATGGC	TGGGTRAARTARGTSACCAGAAYCAGCGG
36	EURL S-13.6	OXA	-48	PCR (published)	Poirel et al. 2004. doi: 10.1128/AAC.48.1.15-22.2004	ttggtggcatcgattatcgg	gagcacttcttttgtgatggc
36	EURL S-13.7	CTX	M-15	PCR (published)	Hasman et al beta-Lactamases among extendedspectrum beta-lactamase (ESBL)-resistant Salmonella from poultry, poultry products and human patients in The Netherlands. JAC 2005 Jul;56(1):115-21	ATGTGCAGYACCAGTAARGTKATGGC	TGGGTRAARTARGTSACCAGAAYCAGCGG
36	EURL S-13.7	SHV	-12	PCR (published)	Briñas et al Beta-lactamases in ampicillinresistant Escherichia coli isolates from foods, humans, and healthy animals. AAC 2002 Oct;46(10):3156-63	CACTCAAGGATGTATTGTG	TTAGCGTTGCCAGTGCTCG
36	EURL S-13.8	CTX	**M-15**	PCR (published)	Hasman et al. beta-Lactamases among extendedspectrum beta-lactamase (ESBL)-resistant Salmonella from poultry, poultry products and human patients in The Netherlands. JAC 2005 Jul;56(1):115-21	ATGTGCAGYACCAGTAARGTKATGGC	TGGGTRAARTARGTSACCAGAAYCAGCGG
58	EURL S-13.2	TEM		PCR (published)	EURL-AR method	URL-AR primer - TEM-all	URL-AR primer - TEM-all
58	EURL S-13.4	CTX		PCR (published)	EURL-AR method	EURL-AR primer CTX-M9	EURL-AR primer CTX-M9
58	EURL S-13.4	CTX		PCR (published)	EURL-AR method	EURL-AR primer - CTX M-all	EURL-AR primer - CTX M-all
58	EURL S-13.4	TEM		PCR (published)	EURL-AR method	EURL-AR primer TEM-all	EURL-AR primer TEM-all
58	EURL S-13.6	OXA		PCR (published)	EURL-AR method	EURL-AR primer - OXA-48	EURL-AR primer - OXA-48
58	EURL S-13.7	CTX		PCR (published)	EURL-AR method	EURL-Ar primer - CTX M-all	EURL-Ar primer - CTX M-all
58	EURL S-13.7	CTX		PCR (published)	EURL-AR method	EURL-Ar primer - CTX-M1	EURL-Ar primer - CTX-M1
58	EURL S-13.7	SHV		PCR (published)	EURL-AR method	EURL-Ar primer - SHV	EURL-Ar primer - SHV
58	EURL S-13.8	**CMY**		PCR (published)	EURL-AR method	EURL-AR primer - CMY-2	EURL-AR primer - CMY-2
58	EURL S-13.8	CTX		PCR (published)	EURL-AR method	EURL-AR primer - CTX-M1	EURL-AR primer - CTX-M1
58	EURL S-13.8	TEM		PCR (published)	EURL-AR method	EURL-AR primer - TEM-all	EURL-AR primer - TEM-all

Legend:

Fields shaded grey indicate that the gene was expected

Genes in bold and white font, were detected but not expected

Note: TEM-1 does not confer ESBL-production and is as such not included as an expected result. TEM-1 or TEM-1B was, however, present in S-13.4 and S-13.8. As S-13.2 was not ESBL, AmpC or carbapenemase producing, submitted resu

Genotypic characterization (optional); comments by participants

Labno	Strain	Comment
17	EURL S-13.8	non ß-lactam resistance genes: aadA2, armA, aac(3)-lid, aac(6')-laa, mph(E), msr(E), sul1, dfrA12; TEM-1B: not ESBL
17	EURL S-13.7	non ß-lactam resistance genes: aph(6)-Id, aph(3'')-Ib, aac(6')-Iic, aac(6')-Iaa, catA2, sul1, tet(D), dfrA19
17	EURL S-13.6	non ß-lactam resistance genes: aac(6')-Iaa, mph(A); chromosomal point
17	EURL S-13.5	non ß-lactam resistance genes: mcr-4.6, aph(6)-Id, aac(6')-Iaa, sul2, dfrA
17	EURL S-13.4	non ß-lactam resistance genes: aac(6')-Iaa, tet(A); chromosomal point mutation gyrA p.S83; TEM-1B: not ESBL
17	EURL S-13.2	non ß-lactam resistance genes: aadA1, aac(6')-Iaa, mph(A), lnu(F); TEM-1B: not ESBL
17	EURL S-13.1	non ß-lactam resistance genes: aadA1, aac(6')-Iaa, sul2, tet(A)
20	EURL S-13.8	blaCTX-M-3 and blaTEM-1B identified using ResFinder
20	EURL S-13.7	blaCTX-M-15 and blaSHV-12 identified using ResFinder
20	EURL S-13.6	blaOXA-48 identified using ResFinder
20	EURL S-13.4	blaCTX-M-9 and blaTEM-1B identified using ResFinder
20	EURL S-13.2	blaTEM-1B identified using ResFinder- not an esbl
20	EURL S-13.1	CTX-M-8 detected through ResFinder
22	EURL S-13.8	Data analysis performed with ResFinder 3.1 data base.
22	EURL S-13.5	Data analysis performed with ResFinder 3.1 data base.
22	EURL S-13.7	Data analysis performed with ResFinder 3.1 data base.
22	EURL S-13.6	Data analysis performed with ResFinder 3.1 data base.
22	EURL S-13.1	Data analysis performed with ResFinder 3.1 data base.
22	EURL S-13.4	Data analysis performed with ResFinder 3.1 data base.

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